ACCESS DB # 366760

FOR OFFICIAL USE ONLY

PLEASE PRINT CLEARLY

Scientific and Technical Information Center

SEARCH REQUEST FORM

Requester's Full Name: Srivastava Examiner # : Date: 6-8-11

Art Unit: Phone Number: 2- Serial Number: 09-484886

Location (Bldg/Room#): (Mailbox #): Results Format Preferred (circle): PAPER DISK

****

To ensure an efficient and quality search, please attach a copy of the cover sheet, claims, and abstract or fill out the following:

Title of Invention:

Inventors (please provide full names):

Earliest Priority Date: 10-8-98

Search Topic:

Please provide a detailed statement of the search topic, and describe as specifically as possible the subject matter to be searched. Include the elected species or structures, keywords, synonyms, acronyms, and registry numbers, and combine with the concept or utility of the invention. Define any terms that may have a special meaning. Give examples or relevant citations, authors, etc., if known.

For Sequence Searches Only Please include all pertinent information (parent, child, divisional, or issued patent numbers) along with the appropriate serial number.

① ex-hypoxic polycythemic mouse, activity of for in vivo testing for recombinant EPO
↓
baculovirus insect cells

② margin of error in determining MW (fractionation) on SDS page

SEARCH PART 1

=> fil medline drugu drugb pascal biotechno wpix ipa biosis lifesci confsci esbio
ntis inis dissabs embase bioeng anabstr scisearch;d que 134; d que 138; d que 163;
s 138,163;fil capl; d que 153; d que 161; d que 157; d que 159; s 153,161
FILE 'MEDLINE' ENTERED AT 15:11:18 ON 15 JUN 2011

FILE 'DRUGU' ENTERED AT 15:11:18 ON 15 JUN 2011
COPYRIGHT (C) 2011 THOMSON REUTERS

FILE 'DRUGB' ENTERED AT 15:11:18 ON 15 JUN 2011
COPYRIGHT (C) 2011 THOMSON REUTERS

FILE 'PASCAL' ENTERED AT 15:11:18 ON 15 JUN 2011
Any reproduction or dissemination in part or in full,
by means of any process and on any support whatsoever
is prohibited without the prior written agreement of INIST-CNRS.
COPYRIGHT (C) 2011 INIST-CNRS. All rights reserved.

FILE 'BIOTECHNO' ENTERED AT 15:11:18 ON 15 JUN 2011
COPYRIGHT (C) 2011 Elsevier Science B.V., Amsterdam. All rights reserved.

FILE 'WPIX' ENTERED AT 15:11:18 ON 15 JUN 2011
COPYRIGHT (C) 2011 THOMSON REUTERS

FILE 'IPA' ENTERED AT 15:11:18 ON 15 JUN 2011
Copyright (c) 2011 The Thomson Corporation

FILE 'BIOSIS' ENTERED AT 15:11:18 ON 15 JUN 2011
Copyright (c) 2011 The Thomson Corporation

FILE 'LIFESCI' ENTERED AT 15:11:18 ON 15 JUN 2011
COPYRIGHT (C) 2011 Cambridge Scientific Abstracts (CSA)

FILE 'CONFSCI' ENTERED AT 15:11:18 ON 15 JUN 2011
COPYRIGHT (C) 2011 Cambridge Scientific Abstracts (CSA)

FILE 'ESBIOBASE' ENTERED AT 15:11:18 ON 15 JUN 2011
COPYRIGHT (C) 2011 Elsevier Science B.V., Amsterdam. All rights reserved.

FILE 'NTIS' ENTERED AT 15:11:18 ON 15 JUN 2011
Compiled and distributed by the NTIS, U.S. Department of Commerce.
It contains copyrighted material.
All rights reserved. (2011)

FILE 'INIS' ACCESS NOT AUTHORIZED

FILE 'DISSABS' ENTERED AT 15:11:18 ON 15 JUN 2011
COPYRIGHT (C) 2011 ProQuest Information and Learning Company; All Rights Reserved.

FILE 'EMBASE' ENTERED AT 15:11:18 ON 15 JUN 2011
Copyright (c) 2011 Elsevier B.V. All rights reserved.

FILE 'BIOENG' ENTERED AT 15:11:18 ON 15 JUN 2011
COPYRIGHT (C) 2011 Cambridge Scientific Abstracts (CSA)

FILE 'ANABSTR' ENTERED AT 15:11:18 ON 15 JUN 2011
COPYRIGHT (c) 2011 THE ROYAL SOCIETY OF CHEMISTRY (RSC)

FILE 'SCISEARCH' ENTERED AT 15:11:18 ON 15 JUN 2011

```
L19         291 SEA (EX HYPOXIC OR EXHYPOXIC)
L20        4165 SEA (POLYCYTHEMIC OR POLY CYTHEMIC)
L21     6672721 SEA MOUSE OR MICE OR MURINE
L22         248 SEA L19 AND L20 AND L21
L24           2 SEA L22 AND PATENT/DT
L26         246 SEA L22 NOT L24
L27         221 SEA L26 AND PY<1999
L28           2 SEA L24 AND (PD<19981008 OR AD<19981008 OR PRD<19981008)
L29         223 SEA (L27 OR L28)
L30      160030 SEA EPO OR ERYTHROPOIETIN
L31       83860 SEA BACULOVIR?
L32       61112 SEA INSECT#(2A) CELL#
L34           0 SEA L29 AND L30 AND (L31 OR L32)



L19         291 SEA (EX HYPOXIC OR EXHYPOXIC)
L20        4165 SEA (POLYCYTHEMIC OR POLY CYTHEMIC)
L21     6672721 SEA MOUSE OR MICE OR MURINE
L22         248 SEA L19 AND L20 AND L21
L24           2 SEA L22 AND PATENT/DT
L26         246 SEA L22 NOT L24
L27         221 SEA L26 AND PY<1999
L28           2 SEA L24 AND (PD<19981008 OR AD<19981008 OR PRD<19981008)
L29         223 SEA (L27 OR L28)
L30      160030 SEA EPO OR ERYTHROPOIETIN
L35     2239817 SEA RECOMB?
L38          15 SEA L29 AND L30 AND L35



L19         291 SEA (EX HYPOXIC OR EXHYPOXIC)
L20        4165 SEA (POLYCYTHEMIC OR POLY CYTHEMIC)
L21     6672721 SEA MOUSE OR MICE OR MURINE
L22         248 SEA L19 AND L20 AND L21
L24           2 SEA L22 AND PATENT/DT
L26         246 SEA L22 NOT L24
L27         221 SEA L26 AND PY<1999
L28           2 SEA L24 AND (PD<19981008 OR AD<19981008 OR PRD<19981008)
L29         223 SEA (L27 OR L28)
L30      160030 SEA EPO OR ERYTHROPOIETIN
L33         203 SEA L29 AND L30
L62     4344294 SEA VIVO
L63          41 SEA L33 AND L62



L65          52 (L38 OR L63)


FILE 'CAPLUS' ENTERED AT 15:11:22 ON 15 JUN 2011
USE IS SUBJECT TO THE TERMS OF YOUR STN CUSTOMER AGREEMENT.
PLEASE SEE "HELP USAGETERMS" FOR DETAILS.
COPYRIGHT (C) 2011 AMERICAN CHEMICAL SOCIETY (ACS)

Copyright of the articles to which records in this database refer is
held by the publishers listed in the PUBLISHER (PB) field (available
```

FILE COVERS 1907 - 15 Jun 2011 VOL 154 ISS 25
FILE LAST UPDATED: 14 Jun 2011 (20110614/ED)
REVISED CLASS FIELDS (/NCL) LAST RELOADED: Apr 2011
USPTO MANUAL OF CLASSIFICATIONS THESAURUS ISSUE DATE: Apr 2011

CAplus now includes complete International Patent Classification (IPC) reclassification data for the fourth quarter of 2010.

CAS Information Use Policies apply and are available at:
http://www.cas.org/legal/infopolicy.html

This file contains CAS Registry Numbers for easy and accurate substance identification.
'OBI' IS DEFAULT SEARCH FIELD FOR 'CAPLUS' FILE

L5 687 SEA FILE=CAPLUS SPE=ON ABB=ON POLYCYTHEMIC/BI
L7 924266 SEA FILE=CAPLUS SPE=ON ABB=ON (MICE OR MOUSE OR MURINE)/BI
L9 360 SEA FILE=CAPLUS SPE=ON ABB=ON L5(3A) L7
L12 73 SEA FILE=CAPLUS SPE=ON ABB=ON (EXHYPOXIC OR EX(A)HYPOXIC)/BI

L13 64 SEA FILE=CAPLUS SPE=ON ABB=ON L9(3A)L12
L41 1 SEA FILE=REGISTRY SPE=ON ABB=ON ERYTHROPOIETIN/CN
L43 15109 SEA FILE=CAPLUS SPE=ON ABB=ON L41
L44 1 SEA FILE=CAPLUS SPE=ON ABB=ON L13 AND PATENT/DT
L45 1 SEA FILE=CAPLUS SPE=ON ABB=ON L13 AND REVIEW/DT
L46 63 SEA FILE=CAPLUS SPE=ON ABB=ON L13 NOT L44
L47 59 SEA FILE=CAPLUS SPE=ON ABB=ON L46 AND PY<1999
L48 1 SEA FILE=CAPLUS SPE=ON ABB=ON L44 AND (PD<19981008 OR AD<19981008 OR PRD<19981008)
L49 60 SEA FILE=CAPLUS SPE=ON ABB=ON (L47 OR L48 OR L45)
L51 225465 SEA FILE=CAPLUS SPE=ON ABB=ON RECOMB?/OBI
L52 1789 SEA FILE=CAPLUS SPE=ON ABB=ON L43(L)L51
L53 2 SEA FILE=CAPLUS SPE=ON ABB=ON L49 AND L52

L5 687 SEA FILE=CAPLUS SPE=ON ABB=ON POLYCYTHEMIC/BI
L7 924266 SEA FILE=CAPLUS SPE=ON ABB=ON (MICE OR MOUSE OR MURINE)/BI
L9 360 SEA FILE=CAPLUS SPE=ON ABB=ON L5(3A) L7
L12 73 SEA FILE=CAPLUS SPE=ON ABB=ON (EXHYPOXIC OR EX(A)HYPOXIC)/BI

L13 64 SEA FILE=CAPLUS SPE=ON ABB=ON L9(3A)L12
L41 1 SEA FILE=REGISTRY SPE=ON ABB=ON ERYTHROPOIETIN/CN
L43 15109 SEA FILE=CAPLUS SPE=ON ABB=ON L41
L44 1 SEA FILE=CAPLUS SPE=ON ABB=ON L13 AND PATENT/DT
L45 1 SEA FILE=CAPLUS SPE=ON ABB=ON L13 AND REVIEW/DT
L46 63 SEA FILE=CAPLUS SPE=ON ABB=ON L13 NOT L44
L47 59 SEA FILE=CAPLUS SPE=ON ABB=ON L46 AND PY<1999
L48 1 SEA FILE=CAPLUS SPE=ON ABB=ON L44 AND (PD<19981008 OR AD<19981008 OR PRD<19981008)
L49 60 SEA FILE=CAPLUS SPE=ON ABB=ON (L47 OR L48 OR L45)

```
L50          52 SEA FILE=CAPLUS SPE=ON  ABB=ON  L43 AND L49
L60      579127 SEA FILE=CAPLUS SPE=ON  ABB=ON  VIVO/BI
L61          11 SEA FILE=CAPLUS SPE=ON  ABB=ON  L50 AND L60



L5          687 SEA FILE=CAPLUS SPE=ON  ABB=ON  POLYCYTHEMIC/BI
L7       924266 SEA FILE=CAPLUS SPE=ON  ABB=ON  (MICE OR MOUSE OR MURINE)/BI
L9          360 SEA FILE=CAPLUS SPE=ON  ABB=ON  L5(3A) L7
L12          73 SEA FILE=CAPLUS SPE=ON  ABB=ON  (EXHYPOXIC OR EX(A)HYPOXIC)/BI

L13          64 SEA FILE=CAPLUS SPE=ON  ABB=ON  L9(3A)L12
L41           1 SEA FILE=REGISTRY SPE=ON  ABB=ON  ERYTHROPOIETIN/CN
L43       15109 SEA FILE=CAPLUS SPE=ON  ABB=ON  L41
L44           1 SEA FILE=CAPLUS SPE=ON  ABB=ON  L13 AND PATENT/DT
L45           1 SEA FILE=CAPLUS SPE=ON  ABB=ON  L13 AND REVIEW/DT
L46          63 SEA FILE=CAPLUS SPE=ON  ABB=ON  L13 NOT L44
L47          59 SEA FILE=CAPLUS SPE=ON  ABB=ON  L46 AND PY<1999
L48           1 SEA FILE=CAPLUS SPE=ON  ABB=ON  L44 AND (PD<19981008 OR
                AD<19981008 OR PRD<19981008)
L49          60 SEA FILE=CAPLUS SPE=ON  ABB=ON  (L47 OR L48 OR L45)
L50          52 SEA FILE=CAPLUS SPE=ON  ABB=ON  L43 AND L49
L54        8016 SEA FILE=CAPLUS SPE=ON  ABB=ON  BACULOVIR?/OBI
L55       11949 SEA FILE=CAPLUS SPE=ON  ABB=ON  (INSECT#(2A)CELL#)/BI
L57           0 SEA FILE=CAPLUS SPE=ON  ABB=ON  L50 AND (L54 OR L55)



L5          687 SEA FILE=CAPLUS SPE=ON  ABB=ON  POLYCYTHEMIC/BI
L7       924266 SEA FILE=CAPLUS SPE=ON  ABB=ON  (MICE OR MOUSE OR MURINE)/BI
L9          360 SEA FILE=CAPLUS SPE=ON  ABB=ON  L5(3A) L7
L12          73 SEA FILE=CAPLUS SPE=ON  ABB=ON  (EXHYPOXIC OR EX(A)HYPOXIC)/BI

L13          64 SEA FILE=CAPLUS SPE=ON  ABB=ON  L9(3A)L12
L41           1 SEA FILE=REGISTRY SPE=ON  ABB=ON  ERYTHROPOIETIN/CN
L43       15109 SEA FILE=CAPLUS SPE=ON  ABB=ON  L41
L44           1 SEA FILE=CAPLUS SPE=ON  ABB=ON  L13 AND PATENT/DT
L45           1 SEA FILE=CAPLUS SPE=ON  ABB=ON  L13 AND REVIEW/DT
L46          63 SEA FILE=CAPLUS SPE=ON  ABB=ON  L13 NOT L44
L47          59 SEA FILE=CAPLUS SPE=ON  ABB=ON  L46 AND PY<1999
L48           1 SEA FILE=CAPLUS SPE=ON  ABB=ON  L44 AND (PD<19981008 OR
                AD<19981008 OR PRD<19981008)
L49          60 SEA FILE=CAPLUS SPE=ON  ABB=ON  (L47 OR L48 OR L45)
L50          52 SEA FILE=CAPLUS SPE=ON  ABB=ON  L43 AND L49
L58        1228 SEA FILE=CAPLUS SPE=ON  ABB=ON  INSECT#/OBI(L)TISSUE/OBI
L59           0 SEA FILE=CAPLUS SPE=ON  ABB=ON  L50 AND L58



L66          13 (L53 OR L61)

=> dup rem l65,l66
FILE 'MEDLINE' ENTERED AT 15:11:33 ON 15 JUN 2011

FILE 'DRUGU' ENTERED AT 15:11:33 ON 15 JUN 2011
COPYRIGHT (C) 2011 THOMSON REUTERS

FILE 'PASCAL' ENTERED AT 15:11:33 ON 15 JUN 2011
Any reproduction or dissemination in part or in full,
by means of any process and on any support whatsoever
```

PROCESSING COMPLETED FOR L65
PROCESSING COMPLETED FOR L66
L67 33 DUP REM L65 L66 (32 DUPLICATES REMOVED)
ANSWERS '1-11' FROM FILE MEDLINE
ANSWERS '12-18' FROM FILE DRUGU
ANSWER '19' FROM FILE PASCAL
ANSWER '20' FROM FILE BIOTECHNO
ANSWER '21' FROM FILE WPIX
ANSWER '22' FROM FILE BIOSIS
ANSWER '23' FROM FILE DISSABS
ANSWERS '24-28' FROM FILE EMBASE
ANSWER '29' FROM FILE ANABSTR
ANSWERS '30-33' FROM FILE CAPLUS

=> d iall 1-20; d ifull 21; d iall 22-29; d ibib ab hitind 30-33;fil hom

L67 ANSWER 1 OF 33 MEDLINE on STN DUPLICATE 1
ACCESSION NUMBER: 1995395358 MEDLINE Full-text
DOCUMENT NUMBER: PubMed ID: 7665979
TITLE: Enhancement of erythropoietin production by selective adenosine A2 receptor agonists in response to hypoxia.
AUTHOR: Ohigashi T; Nakashima J; Aggarwal S; Brookins J; Agrawal K; Fisher J W
CORPORATE SOURCE: Department of Pharmacology, Tulane University School of Medicine, New Orleans, LA 70112, USA.
SOURCE: The Journal of laboratory and clinical medicine, (1995 Sep) Vol. 126, No. 3, pp. 299-306.
Journal code: 0375375. ISSN: 0022-2143. L-ISSN: 0022-2143.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
(RESEARCH SUPPORT, NON-U.S. GOV'T)

LANGUAGE: English
FILE SEGMENT: Abridged Index Medicus Journals; Priority Journals
ENTRY MONTH: 199510
ENTRY DATE: Entered STN: 20 Oct 1995
Last Updated on STN: 3 Feb 1997
Entered Medline: 12 Oct 1995

ABSTRACT:
The purpose of this study was to characterize the effects of two new adenosine A2 agonists, 2-(p-(2-carboxyethyl)phenethyl amino)-5'-N-ethylcarboxamidoadenosine (CGS-21680) and N6-(2(3,5-dimethoxyphenyl)-2-(2-methylphenyl)ethyl)-adenosine (DPMA), on ***erythropoietin*** (EPO) production in vivo and in vitro. Intravenous injections of CGS-21680 (100 to 500 nmol/kg mouse /day) and DPMA (50 to 500 nmol/kg mouse/day) for 4 days produced significant increases in serum levels of EPO in ***polycythemic*** mice. CGS-21680 (10(-7) to 10(-6) mol/L) and DPMA (10(-8) to 10(-5) mol/L) also produced significant increases in medium levels of EPO in a cloned EPO-producing Hep3B hepatocellular carcinoma cell line after 18 hours of incubation in 1% O2. Both compounds also increased cellular cAMP levels significantly in a dose-dependent manner after 1 hour of incubation. A2 receptor binding assays with tritiated CGS-21680 revealed a single type of adenosine receptor binding site on Hep3B cell membranes with a dissociation constant of 132.9 nmol/L and a binding capacity of 270.6 fmol/mg protein. The Ki competition binding values versus tritiated CGS-21680 were 217 nmol/L for CGS-21680 and 86.8 nmol/L for DPMA. These results indicate that adenosine A2 receptor activation amplifies ***EPO*** production in response to hypoxia, both in vivo and in vitro.

CONTROLLED TERM: Check Tags: Female
Adenosine: AD, administration & dosage
*Adenosine: AA, analogs & derivatives
Adenosine: ME, metabolism
Adenosine: PD, pharmacology
Animals
*Anoxia: BL, blood
Binding, Competitive
Carcinoma, Hepatocellular: ME, metabolism
Cyclic AMP: ME, metabolism
*Erythropoietin: BI, biosynthesis
Humans
Kinetics
Liver Neoplasms: ME, metabolism
Mice
Phenethylamines: AD, administration & dosage
Phenethylamines: ME, metabolism
*Phenethylamines: PD, pharmacology
*Polycythemia: BL, blood
*Purinergic P1 Receptor Agonists
Tumor Cells, Cultured

CAS REGISTRY NO.: 11096-26-7 (Erythropoietin); 120225-54-9 (2-(4-(2-carboxyethyl)phenethylamino)-5'-N-ethylcarboxamidoadenosine); 120442-40-2 (CGS 24012); 58-61-7 (Adenosine); 60-92-4 (Cyclic AMP)

CHEMICAL NAME: Phenethylamines; Purinergic P1 Receptor Agonists

L67 ANSWER 2 OF 33 MEDLINE on STN DUPLICATE 2
ACCESSION NUMBER: 1993388899 MEDLINE Full-text
DOCUMENT NUMBER: PubMed ID: 8397229
TITLE: Interaction of nitric oxide and cyclic guanosine 3',5'-monophosphate in erythropoietin production.

AUTHOR: Ohigashi T; Brookins J; Fisher J W
CORPORATE SOURCE: Tulane University School of Medicine, Department of Pharmacology, New Orleans, Louisiana 70112.
SOURCE: The Journal of clinical investigation, (1993 Sep) Vol. 92, No. 3, pp. 1587-91.
Journal code: 7802877. ISSN: 0021-9738. L-ISSN: 0021-9738.
Report No.: NLM-PMC288308.
PUB. COUNTRY: United States
DOCUMENT TYPE: (IN VITRO)
Journal; Article; (JOURNAL ARTICLE)
(RESEARCH SUPPORT, NON-U.S. GOV'T)
LANGUAGE: English
FILE SEGMENT: Abridged Index Medicus Journals; Priority Journals
ENTRY MONTH: 199310
ENTRY DATE: Entered STN: 5 Nov 1993
Last Updated on STN: 3 Feb 1997
Entered Medline: 15 Oct 1993

ABSTRACT:
The present study was designed to investigate whether in vivo and in vitro erythropoietin (EPO) production is modulated by nitric oxide (NO) and cyclic guanosine 3',5'-monophosphate (cGMP). Serum levels of EPO in ex-hypoxic polycythemic ***mice*** were significantly increased after injections of 200 micrograms/kg sodium nitroprusside for 4 d. One injection of NG-nitro-L-arginine methyl ester (L-NAME) produced a significant dose-related decrease in serum levels of ***EPO*** in ex-hypoxic polycythemic ***mice*** in response to hypoxia. When EPO producing Hep3B cells were incubated in 1% O2 for 30 min, cGMP levels in the Hep3B cells were significantly elevated, compared with cells incubated in 20% O2. The elevation of cGMP by hypoxia was inhibited by L-NAME (100 microM). Sodium nitroprusside (10 and 100 microM) and NO (2 microM) also significantly increased cGMP levels in Hep3B cells. L-NAME, LY 83583 (6-Anilino-5,8-quinolinedione, a soluble guanylate cyclase inhibitor), and Rp-8-Bromo-cGMPS (Rp-8-Bromo-guanosine 3',5'-cyclic monophosphothioate, a cGMP-dependent protein kinase inhibitor) significantly inhibited the hypoxia-induced increase in medium levels of ***EPO*** in Hep3B cells. 8-Bromo-cGMPS produced a dose-dependent decrease in ***EPO*** messenger RNA levels in Hep3B cells in response to hypoxia. 8-Bromo-cGMP (10(-3) M) produced significant increases in medium levels of ***EPO*** in Hep3B cell cultures incubated under normoxic conditions, which was enhanced by the phosphodiesterase inhibitor, 3-isobutyl-1-methylxanthine (0.2 mM). These results suggest that NO and cGMP may interact in modulating hypoxic stimulation of EPO production.

CONTROLLED TERM: Check Tags: Female
Animals
Arginine: AA, analogs & derivatives
Arginine: PD, pharmacology
*Cyclic GMP: ME, metabolism
*Erythropoietin: BI, biosynthesis
Humans
Mice
Mice, Inbred C3H
NG-Nitroarginine Methyl Ester
*Nitric Oxide: ME, metabolism
Nitroprusside: PD, pharmacology
Polycythemia: ME, metabolism
Tumor Cells, Cultured
CAS REGISTRY NO.: 10102-43-9 (Nitric Oxide); 11096-26-7 (Erythropoietin); 15078-28-1 (Nitroprusside); 50903-99-6 (NG-Nitroarginine Methyl Ester); 74-79-3 (Arginine); 7665-99-8 (Cyclic GMP)

OS.CITING REF COUNT: 2 There are 2 MEDLINE records that cite this record
MEDLINE REFERENCE COUNT: 26 There are 26 cited references available in MEDLINE for this document.

REFERENCE(S): CITED REFERENCES AVAILABLE IN MEDLINE

(1) Bates, J N; Biochem Pharmacol. 1991 Dec 11, V42 Suppl, PS157-65. MEDLINE
(2) Brezis, M; J Clin Invest. 1991 Aug, V88(2), P390-5. MEDLINE
(3) Brune, B; Eur J Biochem. 1990 Sep 24, V192(3), P683-8. MEDLINE
(4) Butt, E; FEBS Lett. 1990 Apr 9, V263(1), P47-50. MEDLINE
(5) Butt, E; Biochem Pharmacol. 1992 Jun 23, V43(12), P2591-600. MEDLINE
(6) Carmichael, J; Cancer Res. 1987 Feb 15, V47(4), P936-42. MEDLINE
(7) Curran, R D; J Exp Med. 1989 Nov 1, V170(5), P1769-74. MEDLINE
(8) Fisher, J W; Annu Rev Pharmacol Toxicol. 1988, V28, P101-22. MEDLINE
(9) Goldberg, M A; Proc Natl Acad Sci U S A. 1987 Nov, V84(22), P7972-6. MEDLINE
(10) Goldberg, M A; Science. 1988 Dec 9, V242(4884), P1412-5. MEDLINE
(11) Goldberg, M A; Blood. 1991 Jan 15, V77(2), P271-7. MEDLINE
(12) Gray, G A; Br J Pharmacol. 1991 May, V103(1), P1218-24. MEDLINE
(13) Hidaka, H; Biochemistry. 1984 Oct 9, V23(21), P5036-41. MEDLINE
(14) Ignarro, L J; FASEB J. 1989 Jan, V3(1), P31-6. MEDLINE
(15) Klatt, P; Biochem J. 1992 Nov 15, V288 (Pt 1), P15-7. MEDLINE
(16) Malgor, L A; Am J Physiol. 1969 Mar, V216(3), P563-6. MEDLINE
(17) Mason-Garcia, M; Kidney Int. 1990 Nov, V38(5), P969-75. MEDLINE
(18) Menon, N K; Proc Soc Exp Biol Med. 1989 Jul, V191(3), P316-9. MEDLINE
(19) Moncada, S; Pharmacol Rev. 1991 Jun, V43(2), P109-42. MEDLINE
(20) Mulsch, A; Naunyn Schmiedebergs Arch Pharmacol. 1989 Jul, V340(1), P119-25. MEDLINE
(21) Mundel, P; Kidney Int. 1992 Oct, V42(4), P1017-9. MEDLINE
(22) Nakashima, J; J Lab Clin Med. 1992 Mar, V119(3), P306-14. MEDLINE
(23) Nelson, P K; J Pharmacol Exp Ther. 1983 Aug, V226(2), P493-9. MEDLINE
(24) Pohl, U; Am J Physiol. 1989 Jun, V256(6 Pt 2), PH1595-600. MEDLINE
(25) Radermacher, J; Kidney Int. 1992 Jun, V41(6), P1549-59. MEDLINE
(26) Ueno, M; Am J Physiol. 1990 Sep, V259(3 Pt 1), PC427-31. MEDLINE

L67 ANSWER 3 OF 33 MEDLINE on STN DUPLICATE 3

ACCESSION NUMBER:	1990023832 MEDLINE Full-text
DOCUMENT NUMBER:	PubMed ID: 2552819
TITLE:	Enhanced erythropoietin secretion in hepatoblastoma cells in response to hypoxia.
AUTHOR:	Ueno M; Seferynska I; Beckman B; Brookins J; Nakashima J; Fisher J W
CORPORATE SOURCE:	Department of Pharmacology, Tulane University School of Medicine, New Orleans, Louisiana 70112.
CONTRACT NUMBER:	AM-13211 (United States NIADDK NIH HHS)
SOURCE:	The American journal of physiology, (1989 Oct) Vol. 257, No. 4 Pt 1, pp. C743-9. Journal code: 0370511. ISSN: 0002-9513. L-ISSN: 0002-9513.
PUB. COUNTRY:	United States
DOCUMENT TYPE:	Journal; Article; (JOURNAL ARTICLE) (RESEARCH SUPPORT, NON-U.S. GOV'T) (RESEARCH SUPPORT, U.S. GOV'T, P.H.S.)
LANGUAGE:	English
FILE SEGMENT:	Priority Journals
ENTRY MONTH:	198911
ENTRY DATE:	Entered STN: 28 Mar 1990 Last Updated on STN: 3 Feb 1997 Entered Medline: 21 Nov 1989

ABSTRACT:

Erythropoietin (Ep) levels in spent culture media of a Hep G2 human hepatoblastoma cell line were measured by radioimmunoassay (RIA), fetal

mouse liver erythroid colony formation (FMLC), and the ***exhypoxic*** polycythemic mouse assay (EHPCMA). The Hep G2 cells at high density produced approximately 700 mU/ml Ep when measured with the RIA. On the other hand, the Ep levels when assayed in EHPCMA and FMLC were 50 and 2,600 mU/ml, respectively. The bioactivity in FMLC was completely neutralized by an antibody to purified human recombinant Ep, indicating that the erythropoietic activity in the Hep G2 spent culture medium was immunologically equivalent to Ep. Ep levels in the medium from low-density Hep G2 cells in 5% O2 and 1% O2 were 2.5- and 4-fold greater, respectively, than that of 20% O2. In contrast, hyperoxia (40% O2) significantly inhibited Ep production. A significant increase in Ep secretion was also observed when the cells were incubated with cobaltous chloride (2 X 10(-6) -2.5 X 10(-4) M). Tunicamycin (0.5 micrograms/ml), which inhibits N-linked glycosylation, significantly reduced the enhancement of Ep secretion induced by hypoxia (1% O2) without affecting cell growth. Forskolin and cholera toxin, each of which increased the levels of cyclic AMP in the Hep G2 cells by 40-fold, produced a significant (P less than 0.05) further increase in Ep secretion in the presence of hypoxia.(ABSTRACT TRUNCATED AT 250 WORDS)

CONTROLLED TERM: Animals
*Carcinoma, Hepatocellular: SE, secretion
Cell Hypoxia
Cell Line
Cholera Toxin: PD, pharmacology
Colony-Forming Units Assay
Cyclic AMP: AN, analysis
Erythropoietin: PD, pharmacology
*Erythropoietin: SE, secretion
Fetus
Hematopoietic Stem Cells: CY, cytology
Hematopoietic Stem Cells: DE, drug effects
Humans
Kinetics
Liver: CY, cytology
Liver: DE, drug effects
*Liver Neoplasms: SE, secretion
Mice
Radioimmunoassay
*Tumor Cells, Cultured: SE, secretion

CAS REGISTRY NO.: 11096-26-7 (Erythropoietin); 60-92-4 (Cyclic AMP); 9012-63-9 (Cholera Toxin)

OS.CITING REF COUNT: 1 There are 1 MEDLINE records that cite this record

L67 ANSWER 4 OF 33 MEDLINE on STN DUPLICATE 5
ACCESSION NUMBER: 1987209179 MEDLINE Full-text
DOCUMENT NUMBER: PubMed ID: 3577820
TITLE: Erythropoietic factors in plasma from neonatal mice . In vivo studies by the exhypoxic polycythaemic mice assay for erythropoietin.
AUTHOR: Sanengen T; Myhre K; Halvorsen S
SOURCE: Acta physiologica Scandinavica, (1987 Mar) Vol. 129, No. 3, pp. 381-6.
Journal code: 0370362. ISSN: 0001-6772. L-ISSN: 0001-6772.
PUB. COUNTRY: ENGLAND: United Kingdom
DOCUMENT TYPE: (COMPARATIVE STUDY)
Journal; Article; (JOURNAL ARTICLE)
(RESEARCH SUPPORT, NON-U.S. GOV'T)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 198706

ENTRY DATE: Entered STN: 3 Mar 1990
Last Updated on STN: 3 Mar 1990
Entered Medline: 15 Jun 1987

ABSTRACT:
The erythropoiesis stimulating factor(s) (ESF) in plasma from 20-day-old WLO-***mice*** have previously been studied by a cell culture assay, and also by means of gel filtration chromatography and affinity chromatography. It was concluded that the high levels of ESF found in the neonatal mouse plasma probably consisted of erythropoietin (Ep) alone. The objective of the present investigation was to obtain further information of whether this high ESF found in vitro is Ep alone, or Ep in combination with other factors. To accomplish this plasma from 20-day-old WLO mice and a standard Ep were studied in vivo by the exhypoxic ***polycythaemic*** mice assay for Ep, with and without preincubation with rabit anti-Ep serum (AS). Aliquots of some samples were also studied in vitro by the exhypoxic polycythaemic ***mice*** assay for Ep, with and without pre- in both assays (P less than 0.001). However, incubation with AS significantly reduced (P less than 0.001) but did not totally block either the in vivo or the in vitro activity of the plasma (P less than 0.005). This also was the case regarding the in ***vivo*** activity of the standard Ep (P less than 0.001), while the in vitro activity of this Ep preparation was totally blocked by incubation with AS (P greater than 0.3). These results indicate that a considerable part of the high erythropoietic stimulatory activity found in plasma from 20-day-old ***mice*** , with both assays, is Ep. This supports the previous in vitro studies. However, the present results also support the conclusion that part of the activity is due to non-Ep stimulatory factors.

CONTROLLED TERM: Check Tags: Female; Male
Age Factors
Anemia: BL, blood
Animals
Anoxia
*Erythropoiesis
*Erythropoietin: BL, blood
Mice
Mice, Inbred Strains
*Polycythemia: BL, blood
Sheep
Stimulation, Chemical

CAS REGISTRY NO.: 11096-26-7 (Erythropoietin)

L67 ANSWER 5 OF 33 MEDLINE on STN DUPLICATE 6
ACCESSION NUMBER: 1987107668 MEDLINE Full-text
DOCUMENT NUMBER: PubMed ID: 3542810
TITLE: Characterization and biological effects of recombinant human erythropoietin.
AUTHOR: Egrie J C; Strickland T W; Lane J; Aoki K; Cohen A M; Smalling R; Trail G; Lin F K; Browne J K; Hines D K
SOURCE: Immunobiology, (1986 Sep) Vol. 172, No. 3-5, pp. 213-24.
Journal code: 8002742. ISSN: 0171-2985. L-ISSN: 0171-2985.
PUB. COUNTRY: GERMANY, WEST: Germany, Federal Republic of
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 198703
ENTRY DATE: Entered STN: 2 Mar 1990
Last Updated on STN: 2 Mar 1990
Entered Medline: 4 Mar 1987

ABSTRACT:

Human recombinant erythropoietin (rHuEPO) has been purified to apparent homogeneity and compared to purified human urinary ***erythropoietin*** (EPO). Both the purified natural and ***recombinant*** EPO preparations were characterized in a competition radioimmunoassay (RIA), the anhypoxic ***polycythemic*** mouse bioassay, in vitro tissue culture bioassays using bone marrow cells, and by Western analysis. In the immunological and biological activity assays, the rHuEPO shows a dose response which parallels that of the natural hormone. By Western analysis, the ***recombinant*** and human urinary EPO migrate identically. Administration of rHuEPO increases the hematocrit of normal mice in a dose-dependent manner. Additionally, the rHuEPO is able to increase the hematocrit of rats made uremic as a result of subtotal nephrectomy. In summary, by all criteria examined, the rHuEPO is biologically active and equivalent to the natural hormone.

CONTROLLED TERM: Animals
Biological Assay
Bone Marrow Cells
Cells, Cultured
*Erythropoiesis: DE, drug effects
*Erythropoietin: GE, genetics
Erythropoietin: PD, pharmacology
Humans
Immunosorbent Techniques
Mice
Radioimmunoassay
Recombinant Proteins: PD, pharmacology
Uremia: TH, therapy

CAS REGISTRY NO.: 11096-26-7 (Erythropoietin)
CHEMICAL NAME: Recombinant Proteins
OS.CITING REF COUNT: 8 There are 8 MEDLINE records that cite this record

L67 ANSWER 6 OF 33 MEDLINE on STN DUPLICATE 8
ACCESSION NUMBER: 1982109334 MEDLINE Full-text
DOCUMENT NUMBER: PubMed ID: 7324503
TITLE: Effect of membrane dialysis and filtration-sterilization on erythropoietin activity.
AUTHOR: Gallicchio V S; Murphy M J Jr
CONTRACT NUMBER: AM-07266 (United States NIADDK NIH HHS)
AM-19741 (United States NIADDK NIH HHS)
HL-10880 (United States NHLBI NIH HHS)
SOURCE: The Yale journal of biology and medicine, (1981 Jul-Aug) Vol. 54, No. 4, pp. 249-54.
Journal code: 0417414. ISSN: 0044-0086. L-ISSN: 0044-0086.
Report No.: NLM-PMC2595979.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
(RESEARCH SUPPORT, U.S. GOV'T, P.H.S.)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 198203
ENTRY DATE: Entered STN: 17 Mar 1990
Last Updated on STN: 3 Feb 1997
Entered Medline: 22 Mar 1982

ABSTRACT:
Most erythropoietin (Ep) preparations contain non-***erythropoietin*** contaminants. The use of such hormone concentrates raises important questions regarding interpretations of results derived from in ***vivo*** and especially from in vitro studies. By sterilizing various Ep preparations with Nalgene, Millipore, or Selas silver filtration, or even after

conventional membrane dialysis, variable responses were noted when the Ep was assayed with mouse bone marrow cells in vitro (i.e. by stimulating the production of erythroid colonies from CFU-e and BFU-e) and in vivo (i.e., by using the exhypoxic, polycythemic mouse bioassay for Ep). The utility and limitations of such preparative procedures are discussed.

CONTROLLED TERM: Check Tags: Male
Animals
Biological Assay
Bone Marrow: ME, metabolism
Colony-Forming Units Assay
Dialysis
Erythropoietin: IP, isolation & purification
*Erythropoietin: ME, metabolism
Humans
Mice
Sheep
*Sterilization
Ultrafiltration

CAS REGISTRY NO.: 11096-26-7 (Erythropoietin)

OS.CITING REF COUNT: 1 There are 1 MEDLINE records that cite this record

MEDLINE REFERENCE COUNT: 11 There are 11 cited references available in MEDLINE for this document.

REFERENCE(S): CITED REFERENCES AVAILABLE IN MEDLINE

(1) Berman, I; Nature. 1967 Jan 21, V213(5073), P300-1. MEDLINE
(2) Boggs, D R; Blood. 1976 Feb, V47(2), P339-40. MEDLINE
(3) Cahn, R D; Science. 1967 Jan 13, V155(3759), P195-6. MEDLINE
(4) Fisher, J W; Pharmacol Rev. 1972 Sep, V24(3), P459-508. MEDLINE
(5) Gallicchio, V S; Exp Hematol. 1979 May, V7(5), P219-24. MEDLINE
(6) Gordon, A S; Vitam Horm. 1973, V31, P105-74. MEDLINE
(7) Iscove, N N; Exp Hematol. 1975 Jan, V3(1), P32-43. MEDLINE
(8) LOWRY, O H; J Biol Chem. 1951 Nov, V193(1), P265-75. MEDLINE
(9) Lowy, P H; Biochim Biophys Acta. 1968 Aug 13, V160(3), P413-9. MEDLINE
(10) Miyake, T; J Biol Chem. 1977 Aug 10, V252(15), P5558-64. MEDLINE
(11) Shadduck, R K; Exp Hematol. 1978 Apr, V6(4), P355-60. MEDLINE

L67 ANSWER 7 OF 33 MEDLINE on STN DUPLICATE 9

ACCESSION NUMBER: 1983132036 MEDLINE Full-text

DOCUMENT NUMBER: PubMed ID: 6761139

TITLE: Prostaglandins activation of erythropoietin production and erythroid progenitor cells.

AUTHOR: Fisher J W; Radtke H W; Jubiz W; Nelson P K; Burdowski A

CONTRACT NUMBER: AM-13211 (United States NIADDK NIH HHS)
GM-07177 (United States NIGMS NIH HHS)

SOURCE: Experimental hematology, (1980) Vol. 8 Suppl 8, pp. 65-89.
Journal code: 0402313. ISSN: 0301-472X. L-ISSN: 0301-472X.

PUB. COUNTRY: United States

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
(RESEARCH SUPPORT, NON-U.S. GOV'T)
(RESEARCH SUPPORT, U.S. GOV'T, P.H.S.)

LANGUAGE: English

FILE SEGMENT: Priority Journals

ENTRY MONTH: 198304

ENTRY DATE: Entered STN: 18 Mar 1990
Last Updated on STN: 3 Feb 1997
Entered Medline: 15 Apr 1983

ABSTRACT:

A model is presented postulating a role for prostaglandins E and prostacyclin

in kidney generation of erythropoietin and the activation of the erythroid progenitor cell (CFU-E) compartment by erythropoietin (Ep). Several criteria have been met to prove that prostanoids mediate erythropoiesis: 1) several E-type prostaglandins (PGE2, 15-methyl prostaglandin E2, 16,16-dimethyl E2, 6-keto-E1 and PGE1) produced a significant increase in radioiron incorporation in red cells of anypoxic ***polycythemic*** mice; 2) prostaglandin E2 increased kidney production of erythropoietin in the isolated perfused dog kidney; 3) arachidonic acid, a precursor for all bisenoic prostaglandins, increased kidney production of erythropoietin in the isolated perfused dog kidney which was blocked by pretreatment with the cyclo-oxygenase inhibitor drug indomethacin; 4) hypoxemic perfusion of the isolated perfused dog kidney increased kidney production of erythropoietin and produced an elevation in prostacyclin in the perfusates; 5) albuterol, a beta-2 adrenergic agonist, produced a significant increase in perfusate levels of ***erythropoietin*** and PGE in the isolated perfused dog kidney; 6) renal ischemia increased Ep and PGE levels in renal venous plasma which was blocked by pretreatment with indomethacin; 7) prostaglandin E2 and arachidonic acid produced a significant increase in erythroid colonies (CFU-E) in vitro in normal mouse bone marrow; 8) E-type prostaglandins (15-methyl E2) increased in vivo erythroid colony (CFU-E) formation in bone marrows of post-hypoxic polycythemic mice; and 9) injections of 15-methyl E2 daily for six weeks in normal and hypoxic mice produced a significant elevation in the total circulating red cell mass. These studies indicate that hypoxic stimulation of kidney production of ***erythropoietin*** may be related to the generation of prostacyclin (PGI2). On the other hand, albuterol and ischemic (reduction in renal blood flow) stimulation of kidney production of erythropoietin involves prostaglandins of the E type. In addition, E-type prostaglandins were found to enhance the effects of erythropoietin in activating erythroid progenitor cells (CFU-E) in the bone marrow. We postulate from our model that prostaglandins E and prostacyclins are involved in the mechanism of kidney production of erythropoietin as well as the activation of the Ep-responsive cell (ERC) compartment.

CONTROLLED TERM: Check Tags: Female
Albuterol: PD, pharmacology
Animals
Dinoprostone
Dogs
Epoprostenol: PD, pharmacology
*Erythropoiesis: DE, drug effects
*Erythropoietin: BI, biosynthesis
*Hematopoietic Stem Cells: CY, cytology
Hematopoietic Stem Cells: ME, metabolism
Indomethacin: PD, pharmacology
Kidney: DE, drug effects
Kidney: ME, metabolism
Meclofenamic Acid: PD, pharmacology
Mice
Mice, inbred ICR
*Prostaglandins: PD, pharmacology
Prostaglandins E: PD, pharmacology
Stimulation, Chemical

CAS REGISTRY NO.: 11096-26-7 (Erythropoietin); 18559-94-9 (Albuterol); 35121-78-9 (Epoprostenol); 363-24-6 (Dinoprostone); 53-86-1 (Indomethacin); 644-62-2 (Meclofenamic Acid)

CHEMICAL NAME: Prostaglandins; Prostaglandins E

OS.CITING REF COUNT: 1 There are 1 MEDLINE records that cite this record

L67 ANSWER 8 OF 33 MEDLINE on STN DUPLICATE 10
ACCESSION NUMBER: 1980062469 MEDLINE Full-text
DOCUMENT NUMBER: PubMed ID: 507046
TITLE: Chemical modification of nuclear proteins by erythropoietin.
AUTHOR: Spivak J L; Peck L
SOURCE: American journal of hematology, (1979) Vol. 7, No. 1, pp. 45-51.
Journal code: 7610369. ISSN: 0361-8609. L-ISSN: 0361-8609.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
(RESEARCH SUPPORT, U.S. GOV'T, P.H.S.)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 198001
ENTRY DATE: Entered STN: 15 Mar 1990
Last Updated on STN: 15 Mar 1990
Entered Medline: 24 Jan 1980

ABSTRACT:
The spleen of the exhypoxic polycythemic mouse was employed as a model system to study the effect of erythropoietin on enzymes that chemically modify nuclear proteins. At selected time intervals after in vivo administration of erythropoietin, acetyltransferase and methyltransferase activity were measured in nuclei isolated from the spleens of treated mice. In addition, the incorporation of labeled methyl and acetate groups into individual histone proteins was also examined. A 36% increase in nuclear acetyltransferase activity was observed eight hours after administration of ***erythropoietin*** , whereas nuclear methyltransferase activity increased by 42% 24 hours after administration of the hormone. Selective acetylation or methylation of individual histone proteins was not observed, and it is concluded that activation of transcription by erythropoietin is not the result of acetylation or methylation of nuclear proteins.

CONTROLLED TERM: Check Tags: Female
Acetylation
Acetyltransferases
Animals
*Erythropoietin: PD, pharmacology
Histones
Liver: EN, enzymology
Methyltransferases
Mice
*Nucleoproteins: ME, metabolism
Sheep
Spleen: EN, enzymology
Transcription, Genetic
CAS REGISTRY NO.: 11096-26-7 (Erythropoietin)
CHEMICAL NAME: Histones; Nucleoproteins; EC 2.1.1.- (Methyltransferases); EC 2.3.1.- (Acetyltransferases)

L67 ANSWER 9 OF 33 MEDLINE on STN DUPLICATE 11
ACCESSION NUMBER: 1979143920 MEDLINE Full-text
DOCUMENT NUMBER: PubMed ID: 34369
TITLE: Effects of terbutaline, a synthetic beta adrenoceptor agonist, on in vivo erythropoietin production.
AUTHOR: Gross D M; Fisher J W
SOURCE: Archives internationales de pharmacodynamie et de therapie, (1978 Dec) Vol. 236, No. 2, pp. 192-201.
Journal code: 0405353. ISSN: 0301-4533. L-ISSN: 0301-4533.

PUB. COUNTRY: Belgium
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
(RESEARCH SUPPORT, U.S. GOV'T, P.H.S.)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 197905
ENTRY DATE: Entered STN: 15 Mar 1990
Last Updated on STN: 6 Feb 1995
Entered Medline: 23 May 1979

ABSTRACT:
Terbutaline sulfate, a new synthetic beta2-adrenoceptor agonist, was found to produce a dose-related increase in 59Fe-incorporation into newly formed red blood cells of exhypoxic polycythemic mice. This effect was blocked by prior treatment of the polycythemic ***mice*** with the potent beta-adrenoceptor antagonist, DL-propranolol. Terbutaline was also infused (i.v.) (500 microgram/kg/min) into restrained unanesthetized rabbits for a period of 5 hr with constant monitoring of arterial blood pressure and periodic blood Po2, Pco2, and pH analyses. Terbutaline was found to significantly elevate plasma erythropoietin titers in rabbits while producing a slight but nonsignificant decrease in mean blood pressure. Terbutaline did not produce a significant effect upon blood gases or blood pH. These data suggest a possible involvement of beta2-adrenoceptor activation of erythropoietin production.

CONTROLLED TERM: Check Tags: Female
Animals
Blood Gas Analysis
Blood Pressure: DE, drug effects
Epinephrine: BL, blood
Erythropoiesis: DE, drug effects
*Erythropoietin: BI, biosynthesis
Hydrogen-Ion Concentration
Mice
Polycythemia: PP, physiopathology
Propranolol: PD, pharmacology
Rabbits
*Terbutaline: PD, pharmacology
Time Factors
CAS REGISTRY NO.: 11096-26-7 (Erythropoietin); 23031-25-6 (Terbutaline); 51-43-4 (Epinephrine); 525-66-6 (Propranolol)

L67 ANSWER 10 OF 33 MEDLINE on STN
ACCESSION NUMBER: 1996311388 MEDLINE Full-text
DOCUMENT NUMBER: PubMed ID: 8756083
TITLE: Possible role of tumor necrosis factor-alpha in erythropoietic suppression by endotoxin and granulocyte/macrophage colony-stimulating factor.
AUTHOR: Udupa K B; Sharma B G
CORPORATE SOURCE: Education and Clinical Center, V.A. Medical Center, Little Rock, AR 72205, USA.
SOURCE: American journal of hematology, (1996 Jul) Vol. 52, No. 3, pp. 178-83.
Journal code: 7610369. ISSN: 0361-8609. L-ISSN: 0361-8609.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
(RESEARCH SUPPORT, NON-U.S. GOV'T)
(RESEARCH SUPPORT, U.S. GOV'T, NON-P.H.S.)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199612

ENTRY DATE: Entered STN: 28 Jan 1997
Last Updated on STN: 28 Jan 1997
Entered Medline: 11 Dec 1996

ABSTRACT:
Injection of bacterial endotoxin or granulocyte/macrophage colony-stimulating factor (GM-CSF) into exhypoxic polycythemic mice simultaneously with erythropoietin (EPO) suppressed erythroid cell formation, as monitored by 59Fe incorporation into circulating red blood cells. This effect was dose-dependent and time-dependent. GM-CSF did not inhibit erythroid cell formation directly, as the antibody to the GM-CSF did not neutralize the effect of endotoxin, the inducer of GM-CSF. The suppression of both agents could be partially corrected by prior injection of a monoclonal antibody to tumor necrosis factor alpha (anti-TNF alpha). These results indicate that the suppression of EPO-induced erythroid cell formation by endotoxin and GM-CSF was due in part to the production of TNF alpha.

CONTROLLED TERM: Check Tags: Female
Animals
*Endotoxins: PD, pharmacology
Erythrocytes: ME, metabolism
*Erythropoiesis: DE, drug effects
Erythropoietin: PD, pharmacology
*Granulocyte-Macrophage Colony-Stimulating Factor: PD, pharmacology
Injections, Intravenous
Iron: ME, metabolism
Mice
Mice, Inbred Strains
Recombinant Proteins
*Tumor Necrosis Factor-alpha: PH, physiology

CAS REGISTRY NO.: 11096-26-7 (Erythropoietin); 7439-89-6 (Iron); 83869-56-1 (Granulocyte-Macrophage Colony-Stimulating Factor)

CHEMICAL NAME: Endotoxins; Recombinant Proteins; Tumor Necrosis Factor-alpha

L67 ANSWER 11 OF 33 MEDLINE on STN

ACCESSION NUMBER: 1978160506 MEDLINE Full-text
DOCUMENT NUMBER: PubMed ID: 347637
TITLE: Cooperative erythropoietic assay of several steroid metabolites in polycythemic mice.
AUTHOR: Fisher J W; Adamson J W; Camiscoli J F; Fried W; Gordon A S; Schooley J; Zanjani E
SOURCE: Steroids, (1977 Dec) Vol. 30, No. 6, pp. 833-45.
Journal code: 0404536. ISSN: 0039-128X. L-ISSN: 0039-128X.
PUB. COUNTRY: United States
DOCUMENT TYPE: (CLINICAL TRIAL)
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 197806
ENTRY DATE: Entered STN: 14 Mar 1990
Last Updated on STN: 3 Feb 1997
Entered Medline: 28 Jun 1978

ABSTRACT:
A blinded cooperative assay of several androstane and pregnane steroid metabolites has been carried out in order to determine whether 5beta-H derivatives are as active as testosterone in stimulating in vivo erythropoiesis. The steroids tested were: testosterone, 5alpha-dihydrotestosterone, 5beta-dihydrotestosterone,

5beta-pregnane-3,20-dione, 3alpha-dihydroxy-5beta-pregnane-11,20-dione and 3beta-hydroxy-5beta-pregnan-20-one. The incorporation of radioactive iron into newly formed red cells in ***exhypoxic polycythemic ***mice*** was used to compare the effects of the steroids. Testosterone and 5alpha-dihydrotestosterone both produced significant increases in 59Fe incorporation. 5beta-dihydrotestosterone, 5beta-pregnane-3,20-dione, 3alpha-hydroxy-5beta-pregnane-11,20-dione and 3beta-hydroxy-5beta-pregnan-20-one were all devoid of significant erythropoietic activity in polycythemic mice in almost all instances. Thus, under the conditions chosen, this study failed to demonstrate that 5beta-steroids increase radioactive iron incorporation in red cells of ***exhypoxic*** polycythemic mice.

CONTROLLED TERM: Check Tags: Female
Androstanes: ME, metabolism
*Androstanes: PD, pharmacology
Animals
Anoxia: ME, metabolism
Anoxia: PP, physiopathology
Clinical Trials as Topic
Dihydrotestosterone: PD, pharmacology
Double-Blind Method
Erythrocytes: DE, drug effects
Erythrocytes: ME, metabolism
*Erythropoiesis: DE, drug effects
Erythropoietin: PD, pharmacology
Hydroxysteroids: PD, pharmacology
Iron: BL, blood
Mice
*Polycythemia: ME, metabolism
Polycythemia: PP, physiopathology
Pregnanediones: PD, pharmacology
Pregnanes: ME, metabolism
*Pregnanes: PD, pharmacology
Stereoisomerism
Testosterone: PD, pharmacology

CAS REGISTRY NO.: 11096-26-7 (Erythropoietin); 521-18-6 (Dihydrotestosterone); 58-22-0 (Testosterone); 7439-89-6 (Iron)

CHEMICAL NAME: Androstanes; Hydroxysteroids; Pregnanediones; Pregnanes

OS.CITING REF COUNT: 1 There are 1 MEDLINE records that cite this record

L67 ANSWER 12 OF 33 DRUGU COPYRIGHT 2011 THOMSON REUTERS on STN

ACCESSION NUMBER: 1997-04370 DRUGU P G A Full-text

TITLE: Analytical methods for the characterization and quality control of pharmaceutical peptides and proteins, using erythropoietin as an example.

AUTHOR: Gilg D; Riedl B; Zier A; Zimmermann M F

CORPORATE SOURCE: Johnson+Johnson; Cilag-Chemie; Biosyn; Calbiochem-Novabiochem; Swiss-Fed.Inst.Technol.

LOCATION: Schaffhausen, Laufelfingen; Zurich, Switz.; Fellbach, Ger.

SOURCE: Pharm.Acta Helv. (71, No. 6, 383-94, 1996) 4 Fig. 2 Tab. 44 Ref.

CODEN: PAHEAA ISSN: 0031-6865

AVAIL. OF DOC.: R.W.Johnson Pharmaceutical Research Institute, a Division of Cilag AG, Hochstrasse 201, CH-8205 Schaffhausen, Switzerland.

LANGUAGE: English

DOCUMENT TYPE: Journal

ABSTRACT:

Analytical methods for the characterization and quality control of pharmaceutical peptides and proteins are reviewed, using erythropoietin (EPO) as an example. The high complexity of biomacromolecules requires the use not only of physicochemical methodologies, but also of immunochemical and biological techniques for their characterization and quality control.

SECTION HEADING: P Pharmacology
G Galenics
A Analysis

CLASSIF. CODE: 18 Hematological
29 Pharmaceutics
69 Reviews
70 Analysis

CONTROLLED TERM:
REVIEW *FT; IN-VIVO *FT; IN-VITRO *FT; LAB.ANIMAL *FT; CHARACTERIZATION *FT; QUALITY-CONTROL *FT
[01] MAIN-TOPIC *FT; OC *FT; PH *FT
[02] ERYTHROPOIETIN-HUMAN *OC; ERYTHROPOIETIN -HUMAN *PH; HPLC *FT; IR *FT; UV *FT; NMR *FT; MASS *FT; SPECTROMETRY *FT; CIRCULAR-DICHROISM *FT; ELECTROPHORESIS *FT; RADIOIMMUNODET. *FT; ELISA *FT; STRUCT. *FT; PURITY *FT; PH-PK *FT; POTENCY *FT; CHROMATOGRAPHY *FT; OPT.ROTATION *FT; SEROLOGY *FT; ANALYSIS *FT; ENZYME-IMMUNODET. *FT; IMMUNODET. *FT; OC *FT; PH *FT
FIELD AVAIL.: AB; LA; CT
FILE SEGMENT: Literature

L67 ANSWER 13 OF 33 DRUGU COPYRIGHT 2011 THOMSON REUTERS on STN
ACCESSION NUMBER: 1993-15425 DRUGU P Full-text
TITLE: Effects of CGS-21680, a Selective Adenosine A2 Receptor Agonist, on Erythropoietin (EPO) Production.
AUTHOR: Ohigashi T; Brookins J; Fisher J W
LOCATION: New Orleans, Louisiana, United States
SOURCE: Clin.Res. (40, No. 4, 819A, 1992)
CODEN: CLREAS ISSN: 0009-9279
AVAIL. OF DOC.: Department of Pharmacology, Tulane University, New Orleans, LA, U.S.A.
LANGUAGE: English
DOCUMENT TYPE: Journal

ABSTRACT:

CGS-21680 i.v. produced marked increases in serum levels of ***erythropoietin*** (EPO) in exhypoxic ***polycythemic*** mice, when compared with controls after a 4-hr exposure to hypoxia. CGS-21680 produced marked increases in medium levels of ***EPO*** in Hep3B hepatocellular carcinoma cell cultures after 18 hr incubation in a hypoxic atmosphere. Cellular levels of cAMP were also increased after 1 hr incubation. Scatchard analyses of (3H)CGS-21680 binding to membrane preparations of Hep3B cells revealed a single class of binding sites. The Kd value correlated with the ED50 for CGS-21680-stimulated cAMP accumulation in Hep3B cells. Results indicate that adenosine A2 receptor, activated by CGS-21680, is involved in the mediation of EPO production. (congress abstract).

SECTION HEADING: P Pharmacology

CLASSIF. CODE: 18 Hematological
63 Receptors
73 Trial Preparations

CONTROLLED TERM:
[01] CGS-21680 *PH; POLYCYTHEMIA *OC; MARROW-DISEASE *OC; IN-VIVO *FT; I.V. *FT; MOUSE *FT; HYPOXIC *FT; BLOOD-SERUM *FT; CONC. *FT; ERYTHROPOIETIN *FT; IN-VITRO *FT; HEP3B-CELL *FT; CARCINOMA *FT; TUMOR-CELL *FT; CYCLIC-AMP *FT; TRITIUM-LABELED *FT; BINDING *FT; MEMBRANE *FT; PURINERGIC *FT; PURINE-RECEPTOR *FT; INJECTION *FT; LAB.ANIMAL *FT; TISSUE-CULTURE *FT; SUBCELL.STRUCT. *FT; RECEPTOR *FT; TRIAL-PREP. *FT; PURINERGICS *FT; CGS-21680 *RN; PH *FT

FIELD AVAIL.: AB; LA; CT; MPC
FILE SEGMENT: Literature

L67 ANSWER 14 OF 33 DRUGU COPYRIGHT 2011 THOMSON REUTERS on STN
ACCESSION NUMBER: 1990-34145 DRUGU P E Full-text
TITLE: Chemical Modification of Erythropoietin: An Increase in In Vitro Activity by Guanidination.
AUTHOR: Satake R; Kozutsumi H; Takeuchi M; Asano K
CORPORATE SOURCE: Kirin
LOCATION: Gunma, Japan
SOURCE: Biochim.Biophys.Acta P (1038, No. 1, 125-29, 1990) 3 Fig. 2 Tab. 33 Ref. ISSN: 0167-4838
AVAIL. OF DOC.: Pharmaceutical Laboratory, Kirin Brewery Co. Ltd., 1-2-2, Souja-machi, Maebashi, Gunma, 371, Japan.
LANGUAGE: English
DOCUMENT TYPE: Journal

ABSTRACT:

In vitro biological activity of recombinant human ***erythropoietin*** (rHuEPO) was sensitive to modification of the lysine, arginine or tyrosine residues, or the COOH groups. Modifications changing the positive charges of lysine residues to neutral or negative caused a loss in activity, whereas modifications leaving the total number of positive charges unchanged did not affect activity. Guanidinated rHuEPO showed an increase in vitro activity, but amidinated rHuEPO had the same activity as native rHuEPO. The guanidinated derivatives were only about half as active as the native rHuEPO in in vivo exhypoxic polycythemic ***mouse*** bioassay. Guanidino groups, together with their positive charges, may play a role in the interaction between receptors and rHuEPO.

SECTION HEADING: P Pharmacology
E Endocrinology

CLASSIF. CODE: 18 Hematological
38 Structure/Activity
49 Peptide Hormones

CONTROLLED TERM:
[01] ERYTHROPOIETIN *PH; POLYCYTHEMIA *OC; MARROW-DISEASE *OC; RECOMBINANT *FT; HUMAN *FT; IN-VITRO *FT; CHEM. *FT; MODIFICATION *FT; STRUCT.ACT. *FT;

IN-VIVO *FT; MOUSE *FT; LAB.ANIMAL *FT;
ERYTHROPO *RN; PH *FT
FIELD AVAIL.: AB; LA; CT
FILE SEGMENT: Literature

L67 ANSWER 15 OF 33 DRUGU COPYRIGHT 2011 THOMSON REUTERS on STN
ACCESSION NUMBER: 1990-10955 DRUGU P Full-text
TITLE: Relationship Between Sugar Chain Structure and Biological Activity of Recombinant Human Erythropoietin Produced in Chinese Hamster Ovary Cells.
AUTHOR: Takeuchi M; Inoue N; Strickland T W; Kubota M; Wada M; Kobata A
CORPORATE SOURCE: Amgen
LOCATION: Maebashi, Tokyo, Japan; Thousand Oaks, California, United States
SOURCE: Proc.Natl.Acad.Sci.U.S.A. (86, No. 20, 7819-22, 1989) 4 Fig. 1 Tab. 34 Ref.
CODEN: PNASA6 ISSN: 0027-8424
AVAIL. OF DOC.: Pharmaceutical Laboratory, Kirin Brewery, 1-2-2 Soujamachi, Maebashi, Gunma 371, Japan.
LANGUAGE: English
DOCUMENT TYPE: Journal

ABSTRACT:

2 Forms of erythropoietin, EPO-bi and EPO-tetra, were isolated from culture medium of a recombinant Chinese hamster ovary cell line, B8-300, into which the human EPO gene had been introduced. EPO-bi showed only 14% of the in vivo activity in mice but 3 times greater in vitro activity in rat bone marrow cells when compared to recombinant human EPO (REPO). ***EPO*** -tetra had activity comparable to REPO. EPO-bi contained the biantennary N-linked sugar complex type as the major sugar chain while ***EPO*** -tetra and REPO contained the tetraantennary complex type. There was a positive correlation between the ratio of tetraantennary to biantennary oligosaccharides and in vivo activity.

SECTION HEADING: P Pharmacology

CLASSIF. CODE: 18 Hematological

CONTROLLED TERM:
IN-VIVO *FT; IN-VITRO *FT; RAT *FT; MARROW *FT;
MOUSE *FT; CHO-CELL *FT; LAB.ANIMAL *FT;
TISSUE-CULTURE *FT; OVARY *FT
[01] ERYTHROPOIETIN *PH; ERYTHROPO *RN; PH *FT
[02] ERYTHROPOIETIN-HUMAN *PH; RECOMBINANT *FT; ERYTHROPH *RN; PH *FT
FIELD AVAIL.: AB; LA; CT
FILE SEGMENT: Literature

L67 ANSWER 16 OF 33 DRUGU COPYRIGHT 2011 THOMSON REUTERS on STN
ACCESSION NUMBER: 1988-27774 DRUGU A Full-text
TITLE: Evaluation of the Stability of Human Erythropoietin in Samples for Radioimmunoassay
AUTHOR: Eckardt K U; Kurtz A; Hirth P; Scigalla P; Wieczorek L; Bauer C
CORPORATE SOURCE: Boehr.Mannheim

LOCATION: Zurich, Cham, Switzerland
SOURCE: Klin.Wochenschr. (66, No. 6, 241-45, 1988) 3 Fig. 10 Ref.
CODEN: KLWOAZ
AVAIL. OF DOC.: Physiologisches Institut der Universitaet, Zuerich, Switzerland.
LANGUAGE: English
DOCUMENT TYPE: Journal

ABSTRACT:

An evaluation was made of the stability of human recombinant ***erythropoietin*** (ER) in serum and plasma samples obtained from a uremic and a nonuremic anemic patient, for RIA. No signifiant change in the concentration of ER was found in either the serum or plasma samples for up to 14 days of storage, and this stability was observed at a wide range of temperatures. There was no difference between the estimates of ER in serum and heparinized plasma. It was concluded that data obtained clearly indicate that the necessity of storage and transport of clinical samples does not limit the practicability of the RIA for ER.

SECTION HEADING: A Analysis

CLASSIF. CODE: 18 Hematological
70 Analysis

CONTROLLED TERM:
[01] ERYTHROPOIETIN *OC; APLASTIC *OC; ANEMIA *OC; MARROW-DISEASE *OC; NEPHROPATHY *OC; HEPARIN *RC; IN-VITRO *FT; RECOMBINANT *FT; STABILITY *FT; BLOOD-SERUM *FT; BLOOD-PLASMA *FT; RADIOIMMUNODET. *FT; TIME *FT; TEMPERATURE *FT; CASES *FT; ANALYSIS *FT; SEROLOGY *FT; IMMUNODET. *FT; ERYTHROPO *RN; OC *FT
FIELD AVAIL.: AB; LA; CT; MPC
FILE SEGMENT: Literature

L67 ANSWER 17 OF 33 DRUGU COPYRIGHT 2011 THOMSON REUTERS on STN
ACCESSION NUMBER: 1988-37881 DRUGU P E Full-text
TITLE: A1 and A2 Adenosine Receptor Regulation of Erythropoietin Production.
AUTHOR: Ueno M; Brookins J; Beckman B; Fisher J W
LOCATION: New Orleans, Louisiana, United States
SOURCE: Life Sci. (43, No. 3, 229-37, 1988) 4 Fig. 2 Tab. 20 Ref.
CODEN: LIFSAK ISSN: 0024-3205
AVAIL. OF DOC.: Department of Pharmacology, Tulane University School of Medicine, New Orleans, Louisiana 70112, U.S.A.
LANGUAGE: English
DOCUMENT TYPE: Journal

ABSTRACT:

I.v. adenosine hemisulfate (AD) increased the % 59Fe incorporation in RBC of ***exhypoxic*** polycythemic mice. 5'-N-ethyl-carboxamideadenosine (NA) given i.v. increased radioiron incorporation (RI) dose-dependently whereas i.v. N6-cyclohexyladenosine (CA, all Sigma-Chemical) had no effect. I.p. albuterol (AB, Schering-USA) enhanced RI and this enhancement was inhibited by i.v. CA. The AD and NA enhancement was blocked by i.p. theophylline (TH), but was not attenuated by i.p. dipyridamole (DP, both Sigma-Chemical). AD may inhibit, through A1 receptor activation and increase via

A2 receptor stimulation, the production of erythropoietin.

SECTION HEADING: P Pharmacology
E Endocrinology

CLASSIF. CODE: 18 Hematological
49 Peptide Hormones
73 Trial Preparations

CONTROLLED TERM:
MOUSE *FT; IN-VIVO *FT; ERYTHROCYTE *FT; ERYTHROPOIETIN *FT; BIOSYNTH. *FT; HORMONE-METAB. *FT; LAB.ANIMAL *FT
[01] ADENOSINE *PH; SIGMA-CHEM. *FT; SULFATE *PH; I.V. *FT; PURINERGIC *FT; A1 *FT; A2 *FT; INJECTION *FT; PURINERGICS *FT; ADENOSINE *RN; PH *FT
[02] B-744-96 *PH; SIGMA-CHEM. *FT; I.V. *FT; PURINERGIC *FT; A2 *FT; INJECTION *FT; PURINERGICS *FT; CARDIANTS *FT; TRIAL-PREP. *FT; B-744-96 *RN; PH *FT
[03] CYCLOHEXYLADENOSINE *PH; SIGMA-CHEM. *FT; I.V. *FT; PURINERGIC *FT; A1 *FT; INJECTION *FT; PURINERGICS *FT; CYCLOHEAD *RN; PH *FT
[04] SALBUTAMOL *PH; SCHERING-USA *FT; I.P. *FT; SYMPATHOMIMETIC-BETA *FT; BETA-2 *FT; INJECTION *FT; ANTIASTHMATICS *FT; BRONCHODILATORS *FT; SYMPATHOMIMETICS-BETA *FT; TOCOLYTICS *FT; SALBUTAMO *RN; PH *FT
[05] THEOPHYLLINE *PH; SIGMA-CHEM. *FT; I.P. *FT; PURINE-ANTAGONIST *FT; A1 *FT; A2 *FT; INJECTION *FT; BRONCHODILATORS *FT; VASODILATORS *FT; CARDIANTS *FT; DIURETICS *FT; ANTIASTHMATICS *FT; PHOSPHODIESTERASE-INHIBITORS *FT; THEOPHYLL *RN; PH *FT
[06] DIPYRIDAMOLE *PH; SIGMA-CHEM. *FT; I.P. *FT; INJECTION *FT; CARDIANTS *FT; CALCIUM-ANTAGONISTS *FT; ANTIAGGREGANTS *FT; PHOSPHODIESTERASE-INHIBITORS *FT; DIPYRIDAM *RN; PH *FT
FIELD AVAIL.: AB; LA; CT
FILE SEGMENT: Literature

L67 ANSWER 18 OF 33 DRUGU COPYRIGHT 2011 THOMSON REUTERS on STN
ACCESSION NUMBER: 1985-35807 DRUGU P Full-text
TITLE: The Effects of Interferon on Murine Erythropoiesis.
AUTHOR: Huie M L; Gordon A S; Mirand E A; Leong S; Preti R A; Naughton B A
LOCATION: Buffalo, New York New York, United States
SOURCE: Life Sci. (36, No. 26, 2459-62, 1985) 1 Fig. 22 Ref.
CODEN: LIFSAK ISSN: 0024-3205
AVAIL. OF DOC.: New York University, Department of Biology, 100 Washington Square East, New York, New York, 10003, U.S.A.
LANGUAGE: English
DOCUMENT TYPE: Journal

ABSTRACT:

The action of i.p. erythropoietin (EP) in anhypoxic, ***polycythemic*** mice was significantly decreased after low-dose i.m. murine alpha-interferon (IF, Lee-Biomolecular) as assessed by i.p. 59Fe incorporation into RBC. Aditionally, renal EP production in normal intact mice was also significantly decreased following IF and hypoxic exposure. The data suggest that long-term IF treatment may have detrimental

effects on the erythropoietic system both in the responsiveness to and the production of EP.

SECTION HEADING: P Pharmacology

CLASSIF. CODE: 18 Hematological
20 Immunological

CONTROLLED TERM:
HYPOXIA *OC; POLYCYTHEMIA *OC; MARROW-DISEASE *OC; RESPIRATION-DISORDER *OC; MOUSE *FT; IN-VIVO *FT; ERYTHROPOIESIS *FT; ERYTHROCYTE *FT; INJECTION *FT; LAB.ANIMAL *FT
[01] ERYTHROPOIETIN *PH; I.P. *FT; ERYTHROPO *RN; PH *FT
[02] INTERFERON-ALPHA *PH; LEE-BIOMOLECULAR *FT; I.M. *FT; BIOSYNTH. *FT; ERYTHROPOIETIN *FT; VIRUCIDES *FT; IMMUNOSTIMULANTS *FT; CYTOSTATICS *FT; INTERFERA *RN; PH *FT
FIELD AVAIL.: AB; LA; CT
FILE SEGMENT: Literature

L67 ANSWER 19 OF 33 PASCAL COPYRIGHT 2011 INIST-CNRS. ALL RIGHTS RESERVED. on STN
ACCESSION NUMBER: 1994-0067093 PASCAL Full-text
COPYRIGHT NOTICE: Copyright .COPYRGT. 1994 INIST-CNRS. All rights reserved.
TITLE (IN ENGLISH): Effects of 5'-N-ethylcarboxamideadenosine (NECA) on erythropoietin production
AUTHOR: NAKASHIMA J.; OHIGASHI T.; BROOKINS J. W.; BECKMAN B. S.; AGRAWAL K. C.; FISHER J. W.
CORPORATE SOURCE: Tulane univ. school medicine, dep. pharmacology, New Orleans LA 70112, United States
SOURCE: Kidney international, (1993), 44(4), 734-740, 36 refs.
ISSN: 0085-2538 CODEN: KDYIA5
DOCUMENT TYPE: Journal
BIBLIOGRAPHIC LEVEL: Analytic
COUNTRY: United States
LANGUAGE: English
AVAILABILITY: INIST-15906, 354000048187130090
ABSTRACT: he present studies were undertaken to assess the effects of 5'-N-ethylcarboxamideadenosine (NECA), an adenosine analogue, on erythropoietin (Epo) production. NECA (0.05 and 0.1 µmol/kg i.v.) produced significant increases in serum Epo levels (368.8±56.1 and 384.6±45.9 mU/ml, respectively) in exhypoxic polycythemic mice after a four hour exposure to hypoxia when compared with hypoxia controls (133.2±18.2 mU/ml). The hypoxic kidney Epo levels were 46.4±13.4 mU/kg kidney which were significantly higher than normoxic kidney Ep levels (<1.24 mU/kg kidney). Theophylline (20 mg/kg i.p.), an adenosine receptor antagonist, significantly inhibited the stimulatory effects of NECA on serum Epo levels
CLASSIFICATION CODE: 002A18; Life sciences; Biological sciences; Vertebrates physiology, Urinary system
CONTROLLED TERM: Exploration; Treatment; Animal; In vivo; Erythropoietin; Mouse; Analog; Adenosine
BROADER TERM: Rodentia; Mammalia; Vertebrata

L67 ANSWER 20 OF 33 BIOTECHNO COPYRIGHT 2011 Elsevier Science B.V. on STN DUPLICATE
ACCESSION NUMBER: 1989:19263354 BIOTECHNO Full-text
TITLE: Enhanced erythropoietin secretion in hepatoblastoma cells in response to hypoxia

AUTHOR: Ueno M.; Seferynska I.; Beckman B.; Brookins J.; Nakashima J.; Fisher J.W.

CORPORATE SOURCE: Department of Pharmacology, Tulane University School of Medicine, New Orleans, LA 70112, United States.

SOURCE: American Journal of Physiology - Cell Physiology, (1989), 257/4 (26/4) (C743-C749)
CODEN: AJPCDD ISSN: 0002-9513

DOCUMENT TYPE: Journal; Article

COUNTRY: United States

LANGUAGE: English

SUMMARY LANGUAGE: English

ABSTRACT: Erythropoietin (Ep) levels in spent culture media of a Hep G2 human hepatoblastoma cell line were measured by radioimmunoassay (RIA), fetal mouse liver erythroid colony formation (FMLC), and the exhypoxic polycythemic mouse assay (EHPCMA). The Hep G2 cells at high density produced .sim.700 mU/ml Ep when measured with the RIA. On the other hand, the Ep levels when assayed in EHPCMA and FMLC were 50 and 2,600 mU/ml, respectively. The bioactivity in FMLC was completely neutralized by an antibody to purified human recombinant Ep, indicating that the erythropoietic activity in the Hep G2 spent culture medium was immunologically equivalent to Ep. Ep levevls in the medium from low-density Hep G2 cells in 5% O.sub.2 and 1% O.sub.2 were 2.5- and 4-fold greater, respectively, than that of 20% O.sub.2. In contrast, hyperoxia (40% O.sub.2) significantly inhibited Ep production. A significant increase in Ep secretion was also observed when the cells were incubated with cobaltous chloride (2 x 10.sup.-.sup.6-2.5 x 10.sup.-.sup.4 M). Tunicamycin (0.5 μg/ml), which inhibits N-linked glysosylation, significantly reduced the enhancement of Ep secretion induced by hypoxia (1% O.sub.2) without affecting cell growth. Forskolin and cholera toxin, each of which increased the levels of cyclic AMP in the Hep G2 cells by 40-fold, produced a significant ($P < 0.05$) further increase in Ep secretion in the presence of hypoxia. No change in Ep levels in the culture medium occurred when Hep G2 cells were treated with forskolin or cholera toxin under normoxic conditions. In contrast, hypoxia alone failed to increase cyclic AMP levels in the Hep G2 cells. These results indicate that hypoxia produces a significant increase in Ep production by Hep G2 cells through a mechanism that is dependent on normal glycosylation of Ep, whereas hypoxic stimulation of Ep production does not depend on endogenous cyclic AMP accumulation.

CONTROLLED TERM: *adenylate cyclase; *cyclic amp; *erythropoietin; *forskolin; *tunicamycin; *hepatoblastoma; *hypoxia; cholera toxin; cell culture; cell strain hepg2; radioimmunoassay; human cell; human; priority journal

CAS REGISTRY NUMBER: (adenylate cyclase) 9012-42-4; (cyclic amp) 60-92-4; (erythropoietin) 11096-26-7; (forskolin) 66575-29-9; (tunicamycin) 11089-65-9

L67 ANSWER 21 OF 33 WPIX COPYRIGHT 2011 THOMSON REUTERS on STN

ACCESSION NUMBER: 1999-105163 [199909] WPIX
CROSS REFERENCE: 1991-148745; 1995-098764; 1995-284791
TITLE: New isolated erythropoietin isoforms - used for increasing haematocrit levels in mammals
DERWENT CLASS: B04
INVENTOR: STRICKLAND T W
PATENT ASSIGNEE: (AMGE-C) AMGEN INC
COUNTRY COUNT: 1

PATENT INFORMATION:

PATENT NO	KIND	DATE	WEEK	LA	PG	MAIN IPC
US 5856298	A	19990105	(199909)*	EN	26	[9]

APPLICATION DETAILS:

PATENT NO	KIND	APPLICATION	DATE
US 5856298	A CIP of	US 1989-421444	19891013
US 5856298	A Cont of	US 1990-598448	19901012
US 5856298	A Cont of	US 1992-942126	19920908
US 5856298	A	US 1994-334882	19941103

PRIORITY APPLN. INFO: US 1994-334882 19941103
US 1989-421444 19891013
US 1990-598448 19901012
US 1992-942126 19920908

INT. PATENT CLASSIF.:
IPC RECLASSIF.: A61K0038-00 [N,A]; A61K0038-00 [N,C]; C07K0014-435 [I,C]; C07K0014-505 [I,A]
ICO: K61K0038:00; M07K0207:00

BASIC ABSTRACT:

US 5856298 A UPAB: 20050829 An isolated biologically active erythropoietin (EPO) isoform is claimed which has a single isoelectric point and has a specific number of sialic acids per molecule, the number being selected from 1-14, and the isoform being the product of the expression of an exogenous DNA sequence in a non-human eukaryotic host cell. Also claimed are: (1) an EPO consisting of EPO molecules having a single specific number of sialic acids per molecule, the number selected from 1-14, and the molecules being the product of the expression of an exogenous DNA sequence in a non-human eukaryotic host cell; (2) a method of preparing EPO molecules having a predetermined number of sialic acids per molecule, the number selected from 1-14, comprising applying material containing EPO to an ion exchange column and selectively eluting the molecules from the column; (3) a method of preparing EPO molecules having a predetermined number of sialic acids per molecule, the number selected from 1-14, comprising applying material containing EPO to a chromatofocussing column and selectively eluting the molecules from the column.

USE - The isolated EPO isoforms have a defined sialic acid content and biological activity, e.g. the relative in vivo specific activities increase stepwise from isoforms having 5 isoforms having 11 sialic acid residues. The EPOs can be used for increasing haematocrit levels in mammals (claimed).

DOCUMENTATION ABSTRACT:

US5856298

An isolated biologically active erythropoietin (EPO) isoform is claimed which has a single isoelectric point and has a specific number of sialic acids per molecule, the number being

selected from 1-14, and the isoform being the product of the expression of an exogenous DNA sequence in a non-human eukaryotic host cell.

Also claimed are:

(1) an EPO consisting of EPO molecules having a single specific number of sialic acids per molecule, the number selected from 1-14, and the molecules being the product of the expression of an exogenous DNA sequence in a non-human eukaryotic host cell;

(2) a method of preparing EPO molecules having a predetermined number of sialic acids per molecule, the number selected from 1-14, comprising applying material containing EPO to an ion exchange column and selectively eluting the molecules from the column;

(3) a method of preparing EPO molecules having a predetermined number of sialic acids per molecule, the number selected from 1-14, comprising applying material containing EPO to a chromatofocussing column and selectively eluting the molecules from the column.

USE

The isolated EPO isoforms have a defined sialic acid content and biological activity, e.g. the relative in vivo specific activities increase stepwise from isoforms having 5 isoforms having 11 sialic acid residues.

The EPOs can be used for increasing haematocrit levels in mammals (claimed).

EXAMPLE

Recombinant EPO was produced as in US4667016. The different isoforms of EPO were purified by preparative isoelectric focussing in a granulated gel bed.

The sialic acid content was determined by modification of the procedure in J. Biol. Chemical 246, 430, 1971. The sialic acid residues were cleaved from the glycoproteins by hydrolysis with 0.35M sulphuric acid at 80°C for 30 minutes and the solutions were neutralised with NaOH prior to analysis.

In order to estimate the amount of EPO protein present, a Bradford protein assay using recombinant EPO having the amino acid sequence of human EPO as standard was performed using the assay reagents and a micro-method procedure.

BIOLOGICAL DATA

The isoforms isolated were assayed by absorbance at 280 nm, by Bradford protein assay and by RIA for EPO to determine the amount of recombinant EPO present. The ex-hypoxic polycythemic mouse bioassay was used to determine the relative in vivo biological activity, Nature 191, 1065, 1965.

The results showed that the relative in vivo activity of EPO increased as a function of sialic acid content up until isoform 11. Isoforms 11-14 had the same relative in vivo bioactivity.

The greater relative in vivo specific activity of EPO isoforms having more sialic acid is most likely due to a longer circulating half-life of these forms.

Isoforms 9 and 13 were labelled with radioactive I 125 and their rate of clearance in rats was determined.

The half-life in circulation was significantly longer for isoform 13 than for isoform 9.(PHP).

FILE SEGMENT: CPI
MANUAL CODE: CPI: B04-B04D2; B04-N02; B14-F03

NO VALID FORMATS ENTERED FOR FILE 'ANABSTR'
REENTER DISPLAY FORMAT FOR ALL FILES (FILEDEFAULT):all

L67 ANSWER 22 OF 33 BIOSIS COPYRIGHT (c) 2011 The Thomson Corporation on STN DUPLICATE 7
AN 1982:279175 BIOSIS Full-text
DN PREV198274051655; BA74:51655
TI IN-VIVO ACTIVITY OF ASIALO ERYTHROPOIETIN IN COMBINATION WITH ASIALO GLYCO PROTEINS.
AU WEILAND E [Reprint author]; HOPPNER W; BLAEKER F; THORN W
CS DEP PEDIATR MED, UNIV HOSP HAMBURG, MARTINISTR 52, D-2000 HAMBURG 20, FRG
SO Blut, (1982) Vol. 44, No. 3, pp. 173-176.
CODEN: BLUTA9. ISSN: 0006-5242.
DT Article
FS BA
LA ENGLISH
AB In vitro active asialo-erythropoietin has no effect on heme synthesis in vivo. Asialo-glycophorin induces a very low 59Fe-uptake rate in heme in exhypoxic polycythemic mice. The combination of asialo-erythropoietin and asialo-glycophorin or asialo-fetuin induced an activation comparable to the activation by native erythropoietin. The combination of asialo-erythropoietin and tested glycoproteins influence the activity of erythropoesis-stimulating capacity of asialo- erythropoietin.
CC Cytology - Animal 02506
Radiation biology - Radiation and isotope techniques 06504
Biochemistry studies - Proteins, peptides and amino acids 10064
Biochemistry studies - Lipids 10066
Biochemistry studies - Carbohydrates 10068
Biochemistry studies - Minerals 10069
Metabolism - Carbohydrates 13004
Metabolism - Proteins, peptides and amino acids 13012
Blood - General and methods 15001
Blood - Blood cell studies 15004
Blood - Blood, lymphatic and reticuloendothelial pathologies 15006
Blood - Lymphatic tissue and reticuloendothelial system 15008
Endocrine - General 17002
Development and Embryology - Morphogenesis 25508
In vitro cellular and subcellular studies 32600
IT Major Concepts
Blood and Lymphatics (Transport and Circulation); Cell Biology; Development; Endocrine System (Chemical Coordination and Homeostasis); Metabolism
IT Miscellaneous Descriptors
MOUSE FETUIN HEME IRON-59 POLYCYTHEMIA ERYTHROPOIESIS STIMULATING CAPACITY
ORGN Classifier
Muridae 86375
Super Taxa
Rodentia; Mammalia; Vertebrata; Chordata; Animalia
Taxa Notes
Animals, Chordates, Mammals, Nonhuman Vertebrates, Nonhuman Mammals, Rodents, Vertebrates
RN 14875-96-8 (HEME)
14596-12-4 (IRON-59)

L67 ANSWER 23 OF 33 DISSABS COPYRIGHT (C) 2011 ProQuest Information and Learning Company; All Rights Reserved on STN
AN 82:5093 DISSABS Order Number: AAR8214814
TI THE EFFECTS OF PORCINE GASTRIC MUCIN ON ERYTHROPOIETIN PRODUCTION AND THE HEMOPOIETIC INDUCTIVE MICROENVIRONMENT

AU KRUGER, RICHARD EDWARD [PH.D.]
CS NEW YORK UNIVERSITY (0146)
SO Dissertation Abstracts International, (1982) Vol. 43, No. 2B, p. 320. Order No.: AAR8214814. 125 pages.
DT Dissertation
FS DAI
LA English
ED Entered STN: 19921118
Last Updated on STN: 19921118
AB Investigations into the effects of Porcine Gastric Mucin (PGM) on erythropoietin (Ep) production were conducted in three groups of rats: intact, nephrectomized, and hepatectomized/nephrectomized. All three groups received either 1.0 ml of PGM (50 mg/ml) or saline per 100 g of body weight and were exposed to hypoxia (0.4 atm for 6 hours) 1 hour later.
Serum from these animals was assayed for Ep content in the ex -hypoxic, polycythemic mouse. Ep levels in all 3 PGM-treated groups were significantly less than that found for the controls. Histochemical evaluation of the kidney and liver revealed PGM uptake by hepatic Kupffer cells. It was proposed that the PGM induced an alteration in Kupffer cell functioning resulting in a decrease in the production or release of the plasma substrate, required for reaction with the kidney derived enzyme, erythrogenin (Eg) to produce Ep. Reduction in extrarenal Ep levels was considered to result from a similar mechanism involving a reduction in the production and/or release of the Eg, the plasma substrate or Ep itself. PGM was assessed as a potential Hemopoietic Inductive Environmental (HIM) influencing agent, both in vivo and in vitro. PGM addition to Ep stimulated methylcellulose cultures, containing murine femoral marrow cells, resulted in significant decreases in the numbers of early erythroid cells (BFU-E/CFU-E) present, as compared to untreated, Ep control cultures. These effects were attributed to a possible generalized shielding of cell membrane receptors, resulting in cell death. PGM given to mice (1 mg in 0.5 ml alpha medium) intravenously, resulted in increased BFU-E and decreased CFU-E, as compared to alpha medium-treated controls; when the femoral marrow cells of both groups of mice were added to Ep stimulated methylcellulose cultures.
These effects were theorized to have resulted from a PGM-induced alteration in the bone marrow HIM similar to those brought about by certain sulphated acid mucopolysaccharides which tend to stimulate BFU-E and inhibit CFU-E.
CC 0306 BIOLOGY, GENERAL

L67 ANSWER 24 OF 33 EMBASE COPYRIGHT (c) 2011 Elsevier B.V. All rights reserved on STN DUPLICATE 12
AN 0048115689 EMBASE Full-text
TI Incomplete erythropofotin activity in normal pnma component•.
AU Dukes, P.P. (correspondence); Hammond, D.
CS Div. HematoL, Child. Hosp., Los Angeles, CA 90027, United States.
SO PROCSOCEXRBIOLJIED., (1971) Vol. 137, No. 3, pp. 1002-1005.
DT Journal; Article
FS CLASSIC
LA English
SL English
ED Entered STN: Jun 2010
Last Updated on STN: Jun 2010
AB Cohn fractions of normal human plasma were surveyed for erythropoietin activity by an in vivo and two in vitro assay systems. Fractions II + III, II + III W, and especially fraction III, were found to stimulate glucosamine C14 incorporation and heme synthesis of marrow cells in culture. Log dose log

response regression lines of plasma fractions and of an erythropoietin standard were found to be parallel. Only traces of activity could be detected by the exhypoxic polycythemic mouse assay (Fe10). Fraction III from several different sources and species was found to be active in vitro. A human fraction III was shown to have a different specific activity relative to a common erythropoietin standard in the two in vitro assays. Subfractionation of fraction III by extraction procedures demonstrated low stability for the activity measured by the 19Fe heme assay, whereas it was possible to obtain without loss a preparation enriched in the activity stimulating glucosamine incorporation.

CT Medical Descriptors:
assay
bone marrow
bone marrow culture
*bullet
extraction
fractionation
heme synthesis
human
in vitro study
mouse
normal human
plasma
polycythemia
species

CT Drug Descriptors:
erythropoietin
glucosamine
heme
iron

RN (erythropoietin) 11096-26-7

RN CAS Supplied: (GLUCOSAMINE) 3416-24-8; (IRON) 7439-89-6; (HEME) 14875-96-8

L67 ANSWER 25 OF 33 EMBASE COPYRIGHT (c) 2011 Elsevier B.V. All rights reserved on STN DUPLICATE 13

AN 0048299774 EMBASE Full-text

TI Erythropoietin: a complex with different in vivo ac.d in vitro activities.

AU Dukes, P.P. (correspondence); Hammond, D.; Shore, N.A.; Ortega, J.A.

CS Oiv. Hematol., Child. Hosp., Los Angeles, CA, United States.

SO J.LAB.CLIN.MED., (1970) Vol. 76, No. 3, pp. 439-444.

DT Journal; Article

FS CLASSIC

LA English

SL English

ED Entered STN: Jun 2010
Last Updated on STN: Jun 2010

AB Erythropoietin preparations exhibiting the same activity in the exhypoxic polycythemic mouse assay, which quantitates new red cell formation, differ from each other in their ability to stimulate heme synthesis and glucosamine incorporation in bone marrow cells in culture. This suggests that erythropoietin action may result from the separate stimulation by different factors of specific processes of erythroid differentiation.

CT Medical Descriptors:
assay
bone marrow cell
erythrocyte
hematopoiesis
heme synthesis
*in vitro study

mouse
stimulation
CT Drug Descriptors:
*erythropoietin
glucosamine
RN CAS Supplied: (GLUCOSAMINE) 3416-24-8; (ERYTHROPOIETIN) 11096-26-7

L67 ANSWER 26 OF 33 EMBASE COPYRIGHT (c) 2011 Elsevier B.V. All rights reserved on STN
AN 1974073640 EMBASE Full-text
TI Renal mechanism underlying cyclic AMP action on erythropoiesis.
AU Peschle, C.; Rappaport, I.A.; D'Avanzo, A.; et. al.
CS Inst. Med. Pathol., II Fac. Med. Surg., Univ. Naples, Italy.
SO British Journal of Haematology, (1973) Vol. 25, No. 3, pp. 393-398.
ISSN: 0007-1048 CODEN: BJHEAL
DT Journal
FS 037 Drug Literature Index
025 Hematology
005 General Pathology and Pathological Anatomy
023 Nuclear Medicine
028 Urology and Nephrology
030 Clinical and Experimental Pharmacology
LA English
AB Dibutyryl cyclic AMP (dbc AMP) was injected into ex hypoxic polycythaemic mice either alone or with anti erythropoietin (anti Ep) serum. Anti Ep totally abolishes the wave of erythropoiesis evoked by dbc AMP. These results might indicate either that the action of this agent is totally Ep dependent, or that a residual amount of endogenous Ep is necessary to allow dbc AMP to exert a direct effect at the marrow level. The latter mechanism, however, is precluded by experiments indicating that administration of moderate amounts of anti Ep, although abolishing totally the erythroid response to dbc AMP, does not induce complete suppression of endogenous Ep activity and erythropoiesis. Furthermore, a significant rise of Ep plasma level is observed in rats receiving dbc AMP. Since this agent does not apparently modify the kinetics of endogenous Ep, it is postulated that dbc AMP induces a rise in Ep production. This phenomenon, although unmodified in ureter ligated animals, is completely abolished by bilateral nephrectomy. It is therefore concluded that the dbc AMP induces in vivo a stimulatory effect on erythropoiesis via increased production of Ep, via a renal mechanism possibly represented by elevated levels of the renal erythropoietic factor.
CT Medical Descriptors:
*erythrocyte
*erythropoiesis
*hypoxia
intraperitoneal drug administration
*kidney
mouse
*nephrectomy
*polycythemia
*radioactivity
theoretical study
*ureter ligation
CT Drug Descriptors:
*cyclic amp
*erythropoietin
*iron
*iron 59
RN (cyclic AMP) 60-92-4; (erythropoietin) 11096-26-7; (iron 59) 14596-12-4; (iron) 14093-02-8, 53858-86-9, 7439-89-6

L67 ANSWER 27 OF 33 EMBASE COPYRIGHT (c) 2011 Elsevier B.V. All rights reserved on STN
AN 0048637092 EMBASE Full-text
TI Control of erythropoiesis in rats with adjuvant induced chronic inflammation.
AU Lukens, J.N. (correspondence)
CS Dept. Ped., Univ. Missouri Sch. Med., Columbia, MO 65201, United States.
SO Blood, (1973) Vol. 41, No. 1, pp. 37-44.
ISSN: 0006-4971
DT Journal; Article
FS CLASSIC
LA English
SL English
ED Entered STN: Jun 2010
Last Updated on STN: Jun 2010
AB In order to characterize the defect in erythroid homeostasis in chronic inflammatory states, the relation between erythropoietin production and erythropoietic response was examined in rats with adjuvant disease. Exposure of adjuvant injected rats to graded levels of lowered barometric pressure induced increases in plasma erythropoietin which were significantly less than those measured in normal animals similarly stimulated. Erythropoietin inhibitors were not detected by in vitro or in vivo assay techniques: the biological activity of ovine erythropoietin was not modified by incubation with plasma from rats with adjuvant disease; the erythropoietic response of ex hypoxic polycythemic mice to erythropoietin was not compromised by injections of test plasma; and the burst of erythropoiesis induced in eshypoxic polycythemic mice by a hypobaric stimulus was not modified by plasma given prior to or at various intervals after hypobaric exposure. Exogenous erythropoietin elicited nearly identical increases of radioiron incorporation in normal and adjuvant injected rats whose endogenous erythropoietin was suppressed by hypertransfusion. It is concluded that the diminished erythropoietic response to anemia in adjuvant induced chronic inflammation results from a relative failure in the production of biologically active erythropoietin.
CT Medical Descriptors:
adjuvant disease
anemia
assay
biological activity
bone marrow
*chronic inflammation
*erythropoiesis
exposure
homeostasis
in vitro study
injection
mouse
plasma
*rat
stimulus
CT Drug Descriptors:
*adjuvant
erythropoietin
Freund adjuvant
RN (erythropoietin) 11096-26-7; (Freund adjuvant) 9007-81-2

L67 ANSWER 28 OF 33 EMBASE COPYRIGHT (c) 2011 Elsevier B.V. All rights reserved on STN
AN 0048115684 EMBASE Full-text

TI T. differences between in vivo end in vitro activitess of various erythropoitin preparations dukes ppn hammond dn.
AU Shore, N.A. (correspondence); Ortega, J.A.
CS Div. Hematol Child. Hosps, Los Angeles.
SO KRABL JJIEDSCL, (1971) Vol. 7, No. 7-8, pp. 919-926.
DT Journal; Article
FS CLASSIC
LA English
SL English
ED Entered STN: Jun 2010
Last Updated on STN: Jun 2010
AB It was found that erythropoietin preparations exhibiting the same activity in the exhypoxic polycythemic mouse assay, which quantitates new red cell formation in vivo, differed from each other in their ability to stimulate heme synthesis and glucosamine incorporation in bone marrow cells in culture. By Chromatographie fractionation of a preparation, it was possible to enrich to a widely different extent activities measured by the three assay systems. This suggests that erythropoietin action may result from the separate stimulation by different factors of specific processes of erythroid differentiation. Alternatively, the presence in the preparations of various inhibitor* of these processes could be the cause of the observed differences in specific activities.
CT Medical Descriptors:
assay
bone marrow cell
erythrocyte
fractionation
heme synthesis
*in vitro study
mouse
stimulation
CT Drug Descriptors:
erythropoietin
glucosamine
RN CAS Supplied: (GLUCOSAMINE) 3416-24-8; (ERYTHROPOIETIN) 11096-26-7

L67 ANSWER 29 OF 33 ANABSTR COPYRIGHT 2011 RSC on STN
AN 59(5):F80 ANABSTR Full-text
TI Erythropoietin: physico- and biochemical analysis.
AU Choi, D.; Kim, M.; Park, J. (Doping Control Center, Korea Inst. Sci. Technol., Seoul 130-650, South Korea)
SO J. Chromatogr., B: Biomed. Appl. (1996) 687(1), 189-199
CODEN: JCBBEP ISSN: 0378-4347
DT Journal
LA English
AB A review is presented on erythropoietin and its possible misuse by athletes. Both the physiological and biochemical characteristics of the hormone are discussed along with purification and analytical methodologies. Techniques, such as, the exhypoxic polycythemic mouse assay, RIA and reticulocyte counts, peptide mapping, carbohydrate microheterogeneity and comparative analysis of natural hormone versus recombinant human hormone are considered. (83 references).
CC *F Clinical and Biochemical Analysis (40000)
IT Matrix:
11096-26-7, erythropoietin
(analysis of, review)

L67 ANSWER 30 OF 33 CAPLUS COPYRIGHT 2011 ACS on STN
ACCESSION NUMBER: 1988:453211 CAPLUS Full-text
DOCUMENT NUMBER: 109:53211
ORIGINAL REFERENCE NO.: 109:8959a,8962a
TITLE: Human erythropoietin gene: high level expression in stably transfected mammalian cells
INVENTOR(S): Powell, Jerry S.
PATENT ASSIGNEE(S): University of Washington, USA
SOURCE: PCT Int. Appl., 22 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE	
WO 8800241	A1	19880114	WO 1987-US1459	19870623	<--
W: FI, KR, LK, MC, MG, MW, NO, RO, SD, SE, SU					
RW: BJ, CF, CG, CM, GA, ML, MR, SN, TD, TG					
DK 8703093	A	19871228	DK 1987-3093	19870617	<--
DK 173067	B1	19991213			
CA 1341361	C	20020521	CA 1987-616544	19870622	<--
EP 255231	A1	19880203	EP 1987-305672	19870625	<--
EP 255231	B1	19920520			
R: AT, BE, CH, DE, ES, FR, GB, GR, IT, LI, LU, NL, SE					
AT 76431	T	19920615	AT 1987-305672	19870625	<--
ES 2037083	T3	19930616	ES 1987-305672	19870625	<--
AU 8774757	A	19880107	AU 1987-74757	19870626	<--
AU 611088	B2	19910606			
BR 8703269	A	19880315	BR 1987-3269	19870626	<--
CN 87104424	A	19880427	CN 1987-104424	19870626	<--
CN 1044133	C	19990714			
CN 1224726	A	19990804	CN 1998-115963	19870626	<--
CN 101041819	A	20070926	CN 2006-10100687	19870626	<--
JP 63126488	A	19880530	JP 1987-160799	19870627	<--
KR 9709935	B1	19970619	KR 1987-6561	19870627	<--
FI 8800899	A	19880226	FI 1988-899	19880226	<--
FI 95393	B	19951013			
FI 95393	C	19960125			
NO 8800863	A	19880426	NO 1988-863	19880226	<--
NO 303398	B1	19980706			
US 5688679	A	19971118	US 1993-132489	19931006	<--
US 20020045255	A1	20020418	US 2001-975063	20011010	<--
US 6867020	B2	20050315			
US 20020137145	A1	20020926	US 2001-11858	20011105	<--
US 6682910	B2	20040127			

PRIORITY APPLN. INFO.:

US 1986-879423	A	19860627	<--
WO 1987-US1459	A	19870623	<--
EP 1987-305672	A	19870625	<--
CN 1987-104424	A3	19870626	<--
CN 1998-115963	A3	19870626	<--
US 1988-211278	B1	19880621	<--
US 1989-453381	B1	19891218	<--
US 1993-132489	A	19931006	<--
US 1995-466412	A1	19950606	<--
US 1999-238055	A1	19990127	

AB Plasmids containing the ApaI fragment of the human erythropoietin (I) gene are constructed and used to stably transfect mammalian cells. These cells secrete high levels of I into the culture medium. Plasmid pBD-EP, containing the ApaI

fragment of the I gene, the MT-1 promoter sequence, the SV40 enhancer, and the dihydrofolate reductase gene, was constructed. BHK cells were transfected with this plasmid and the stable transformants were selected for in methotrexate medium. One such clone produced 6728 units I/mL culture supernatant. I was assayed in vitro (formation of erythroid colonies in mouse bone marrow cell cultures, and by competitive RIA) and in vivo (exhypoxic polycythemic mice).
IPCI C12N0015-00 [ICM,4]; C12N0001-00 [ICS,4]; C12P0021-02 [ICS,4]; C07K0013-00 [ICS,4]
IPCR C12N0015-09 [I,A]; C07H0021-02 [I,A]; C07K0014-00 [I,A]; C07K0014-505 [I,A]; C07K0014-52 [I,A]; C12N0001-16 [I,A]; C12N0001-20 [I,A]; C12N0005-10 [I,A]; C12N0015-86 [I,A]; C12P0021-02 [I,A]; C12R0001-01 [N,A]; C12R0001-645 [N,A]; C12R0001-91 [N,A]
CC 16-2 (Fermentation and Bioindustrial Chemistry)
IT 11096-26-7P, Erythropoietin
RL: BMF (Bioindustrial manufacture); BIOL (Biological study); PREP (Preparation)
(manufacture of, high-level, stably transformed mammalian cells for)
OS.CITING REF COUNT: 15 THERE ARE 15 CAPLUS RECORDS THAT CITE THIS RECORD (18 CITINGS)
REFERENCE COUNT: 5 THERE ARE 5 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L67 ANSWER 31 OF 33 CAPLUS COPYRIGHT 2011 ACS on STN
ACCESSION NUMBER: 1989:51365 CAPLUS Full-text
DOCUMENT NUMBER: 110:51365
ORIGINAL REFERENCE NO.: 110:8325a,8328a
TITLE: Comparison of recombinant and human erythropoietin as antigen in the radioimmunoassay
AUTHOR(S): Mason-Garcia, Meredith; Brookins, Jesse W.; Beckman, Barbara S.; Fisher, James W.
CORPORATE SOURCE: Sch. Med., Tulane Univ., New Orleans, LA, 70112, USA
SOURCE: Journal of Clinical Immunoassay (1988), 11(3), 135-40
CODEN: JCLIES; ISSN: 0736-4393
DOCUMENT TYPE: Journal
LANGUAGE: English
AB RIAs based on the use of highly purified human urinary erythropoietin (huEp) and recombinant human Ep (rEp) were compared with regard to sensitivity, specificity, precision, and correlation with the exhypoxic-polycythemic mouse bioassay. The Ep levels in the sera of normal adults were not significantly different using the huEp or rEp RIAs, and both systems yielded Ep values in the sera of aplastic anemia patients that correlated well with each other and with the exhypoxic-polycythemic mouse bioassay. The dose-response regression lines of diluted standard Ep and diluted serum were parallel in both systems, and the diluted standard huEp and rEp regression lines were superimposable within both the huEp and rEp assays. Thus, these studies provide good evidence that these antigens are immunol. similar and that the standardization of both antigens is equivalent However, several differences were found in these RIA systems, most of which seem to be attributable to variations in the immunoreactivity of the radioiodinated antigens. Although some differences do exist between the rEp and huEp RIAs, results of the rEp assay correlate well with those of the huEp RIA and of the bioassay, and the rEp RIA may be used with confidence for both clin. and research applications.
CC 2-1 (Mammalian Hormones)
IT 11096-26-7, Erythropoietin
RL: BIOL (Biological study)
(human urinary and recombinant, as antigen for RIA)
OS.CITING REF COUNT: 1 THERE ARE 1 CAPLUS RECORDS THAT CITE THIS RECORD (1 CITINGS)

L67 ANSWER 32 OF 33 CAPLUS COPYRIGHT 2011 ACS on STN
ACCESSION NUMBER: 1978:613223 CAPLUS Full-text
DOCUMENT NUMBER: 89:213223
ORIGINAL REFERENCE NO.: 89:33119a,33122a
TITLE: A factor from urine which modulates in vivo erythropoietin activity
AUTHOR(S): Dukes, Peter P.; Ortega, Jorge A.; Shore, Nomie A.; Harris, Kathryn; Polk, Curtiss
CORPORATE SOURCE: Div. Hematol.-Oncol., Child. Hosp. Los Angeles, Los Angeles, CA, USA
SOURCE: Haematologica (1978), 63(4), 420-5
CODEN: HAEMAX; ISSN: 0390-6078
DOCUMENT TYPE: Journal
LANGUAGE: English
AB A protein factor from human anemic urine was separated from erythropoietin by chromatog. on QAE-Sephadex. It stimulated 59Fe incorporation in the exhypoxic polycythemic mouse assay but with characteristics different from those of erythropoietin. Simultaneous injection of fixed amts. of this factor with various erythropoietin doses used to generate dose-response curves led to increases of the responses to small doses but had no effect on or actually decreased the response to larger doses of erythropoietin.
CC 14-9 (Mammalian Pathological Biochemistry)
IT 11096-26-7
RL: PROC (Process)
(protein of urine modulation of)

L67 ANSWER 33 OF 33 CAPLUS COPYRIGHT 2011 ACS on STN
ACCESSION NUMBER: 1974:400944 CAPLUS Full-text
DOCUMENT NUMBER: 81:944
ORIGINAL REFERENCE NO.: 81:159a,162a
TITLE: Differences between in vivo and in vitro activities of various erythropoietin preparations
AUTHOR(S): Dukes, Peter P.; Hammond, Denman; Shore, Nomie A.; Ortega, Jorge A.
CORPORATE SOURCE: Div. Hematol., Child. Hosp., Los Angeles, CA, USA
SOURCE: Erythropoiesis: Regul. Mech. Develop. Aspects, Proc. Tel Aviv Univ. Conf. (1971), Meeting Date 1970, 97-104. Editor(s): Matoth, Yahuda. Academic: New York, N. Y.
CODEN: 28GEAS
DOCUMENT TYPE: Conference
LANGUAGE: English
AB Erythropoietin prepns. exhibiting the same activity in the ex- hypoxic polycythemic mouse assay, which quantitates new erythrocyte formation in vivo, differed from each other in their ability to stimulate heme synthesis and glucosamine incorporation in bone marrow cells in culture. By chromatog. fractionation of a preparation, it was possible to enrich to a widely different extent activities measured by the 3 assay systems. Thus, erythropoietin action may result from the sep. stimulation by different factors of specific processes of erythroid differentiation. Alternatively, the presence in the prepns. of various inhibitors of these processes could be the cause of the observed differences in specific activities.
CC 9-2 (Biochemical Methods)
Section cross-reference(s): 2
IT 11096-26-7
RL: ANT (Analyte); ANST (Analytical study)
(activity determination of, in vivo and vitro)

FILE 'HOME' ENTERED AT 15:12:17 ON 15 JUN 2011

SEARCH PART 2

```
=> fil USPATFULL, PCTFULL, USPAT2, EPFULL, FRFULL, GBFULL
FILE 'USPATFULL' ENTERED AT 15:26:34 ON 15 JUN 2011
CA INDEXING COPYRIGHT (C) 2011 AMERICAN CHEMICAL SOCIETY (ACS)

FILE 'PCTFULL' ENTERED AT 15:26:34 ON 15 JUN 2011
COPYRIGHT (C) 2011 LexisNexis Univentio B.V.

FILE 'USPAT2' ENTERED AT 15:26:34 ON 15 JUN 2011
CA INDEXING COPYRIGHT (C) 2011 AMERICAN CHEMICAL SOCIETY (ACS)

FILE 'EPFULL' ENTERED AT 15:26:34 ON 15 JUN 2011
COPYRIGHT (C) 2011 European Patent Office / FIZ Karlsruhe / LexisNexis Univentio
B.V.

FILE 'FRFULL' ENTERED AT 15:26:34 ON 15 JUN 2011
COPYRIGHT (C) 2011 LexisNexis Univentio B.V.

FILE 'GBFULL' ENTERED AT 15:26:34 ON 15 JUN 2011
COPYRIGHT (C) 2011 LexisNexis Univentio B.V.

=> d que l76; d que l78;d que l71; d que l80; s l76,l78
L69       18198 SEA MARGIN#(1W) ERROR
L70      317803 SEA SDSPAGE OR SDS OR SODIUM DODECYL? OR SODIUMDODECYL?
L73     1073173 SEA MOLECULAR WEIGHT
L74      433002 SEA MW OR M(W) W
L75          21 SEA L69(8A) (L73 OR L74)
L76           4 SEA L75 AND L70


L69       18198 SEA MARGIN#(1W) ERROR
L70      317803 SEA SDSPAGE OR SDS OR SODIUM DODECYL? OR SODIUMDODECYL?
L77      243292 SEA KDA OR KILODALTON# OR DALTON#
L78           8 SEA L69(8A) L77 AND L70


L69       18198 SEA MARGIN#(1W) ERROR
L70      317803 SEA SDSPAGE OR SDS OR SODIUM DODECYL? OR SODIUMDODECYL?
L71           0 SEA L69(5A) L70


L69       18198 SEA MARGIN#(1W) ERROR
L70      317803 SEA SDSPAGE OR SDS OR SODIUM DODECYL? OR SODIUMDODECYL?
L79      251969 SEA FRACTIONAT?
L80           0 SEA L69(8A) L79 AND L70


L82           9 (L76 OR L78)

=> dup rem l82
PROCESSING COMPLETED FOR L82
L83           9 DUP REM L82 (0 DUPLICATES REMOVED)
                ANSWERS '1-2' FROM FILE USPATFULL
                ANSWERS '3-6' FROM FILE PCTFULL
                ANSWER '7' FROM FILE EPFULL
                ANSWERS '8-9' FROM FILE FRFULL

=> d ibib ab kwic l-9; fil hom
```

L83 ANSWER 1 OF 9 USPATFULL on STN
ACCESSION NUMBER: 2002:325994 USPATFULL Full-text
TITLE: Syndecan enhancer element and syndecan stimulation of cellular differentiation
INVENTOR(S): Jalkanen, Markku, Piispanristi, FINLAND
Jaakkola, Panu, Turku, FINLAND
Vihinen, Tapani, Turku, FINLAND
PATENT ASSIGNEE(S): Biotie Therapies Corp., Turku, FINLAND (non-U.S. corporation)

	NUMBER	KIND	DATE
PATENT INFORMATION:	US 6492344	B1	20021210
APPLICATION INFO.:	US 1999-336757		19990621 (9)

RELATED APPLN. INFO.: Continuation-in-part of Ser. No. US 1994-206186, filed on 7 Mar 1994, now abandoned Continuation-in-part of Ser. No. WO 1993-FI514, filed on 1 Dec 1993
DOCUMENT TYPE: Utility
FILE SEGMENT: GRANTED
PRIMARY EXAMINER: Nguyen, Dave T.
ASSISTANT EXAMINER: Shukla, Ram R.
LEGAL REPRESENTATIVE: Sterne, Kessler, Goldstein & Fox P.L.L.C.
NUMBER OF CLAIMS: 11
EXEMPLARY CLAIM: 1
NUMBER OF DRAWINGS: 67 Drawing Figure(s); 47 Drawing Page(s)
LINE COUNT: 2869
CAS INDEXING IS AVAILABLE FOR THIS PATENT.

AB Methods are provided for altering levels of syndecan within a cell. The methods include enhancing syndecan expression via administration of growth factors, preventing suppression of syndecan expression via administration of anti-steroid agents, and altering syndecan biochemistry within the cell. The methods are used to induce or maintain cellular differentiation, and to decrease the growth of malignant cells. Application of the methods to the treatment of patients, including humans, is provided. A syndecan enhancer element, novel proteins that activate the enhancer element, non-human transgenic animals comprising this enhancer element linked to a structural gene, and the use of this enhancer element to regulate the expression of syndecan and other genes are also provided. The enhancer element can also be used to target expression of a gene to wound sites. DRWD . . . produce a supershift with labelled motif 3 and nuclear extracts
deemed from FGF-2 treated 3T3 NIH cells. A gel retardation gel as shown in FIG. 15a was run and exposed to UV light. The specific bands, representing the bound protein-DNA complex, were cut out, eluted overnight, and loaded onto an SDS-PAGE gel to analyze their molecular weight. Two reproducible bands for the motif 3 binding protein are shown. The molecular weight of the nuclear factors were approximated by subtracting the calculated molecular weight of each oligonucleotide from the complex molecular weight.
DETD The FIN-1 protein has been isolated and has a molecular weight of 50 kDa as determined by SDS-PAGE.
DETD SDS-PAGE and Western Blot--For western blot experiments, cells were cultured 24 hours with or without growth factor(s). Syndecan ectodomain containing material released from the cell surface by trypsin treatment was fractionated on SDS-PAGE gradient (2-15%) gel (O'Farrel, J. Biol. Chem. 250:4007-4021 (1975)). After electrophoresis, samples were transferred onto a Zeta-Probe membrane by electroblotting with a 2005 Transphor apparatus (LKB). The syndecan antigen on the filter was detected with radioiodinated mAB 281-2 and the filter was washed, as described above for slot blot analysis.
DETD . . . samples were size-separated on a 1% agarose formaldehyde gel, transferred to a GeneScreen Plus.TM. membrane (New England Nuclear) and

hybridized with a multi-prime (Amersham) labeled partial cDNA clone for mouse syndecan (PM-4) (Saunders et al., J. Cell Biol. 108:1547-1556 (1989)). After hybridization, the membrane was washed in 2+SSC and 1.0% SDS at 65° C. (high stringency conditions). For rehybridization with glyceraldehyde-3-phosphate-dehydrogenase (GAPDH; Fort et al., Nucleic Acid Res. 13:1431-1442 (1985)), the bound PM-4 probe was removed as recommended by the manufacturer of the filter (NEN).

DETD . . . run, it was exposed to 245 nm UV-light (3600J/em.sup.2) in a Strategene crosslinker. The gel was exposed for several hours, the specific bands were cut out, eluted overnight at 4° C., precipitated with ethanol, resuspended in Laemmli buffer, denatured at 95° C. for 5 minutes, and loaded onto a 10% SDS-PAGE together with a .sup.14C-labeled molecular weight markers to analyze their molecular weights. The SDS-PAGE gel is shown in FIG. 15c, with the position of the molecular weight markers shown at the left. Lanes 1-5 correspond to motifs 1-5, respectively. The molecular weights of the nuclear factors were estimated after subtracting the mass of the oligonucleotide from the complex mass as indicated below:

DETD This experiment shows a reproducible 46 kDa band for motif 1 and two bands, 78 kDa and 50 kDa, for motif 3. These values have a margin of error of about ±3 kDa.

L83 ANSWER 2 OF 9 USPATFULL on STN

ACCESSION NUMBER:	2000:9716 USPATFULL Full-text
TITLE:	Syndecan enhancer element and syndecan stimulation of cellular differentiation
INVENTOR(S):	Jalkanen, Markku, Piispanristi, Finland Jaakkola, Panu, Turku, Finland Vihinen, Tapani, Turku, Finland
PATENT ASSIGNEE(S):	BioTie Therapies Ltd., Turku, Finland (non-U.S. corporation)

	NUMBER	KIND	DATE
PATENT INFORMATION:	US 6017727		20000125
APPLICATION INFO.:	US 1996-760534		19961202 (8)
RELATED APPLN. INFO.:	Continuation-in-part of Ser. No. US 1994-206186, filed on 7 Mar 1994, now abandoned which is a continuation-in-part of Ser. No. WO 1993-FI514, filed on 1 Dec 1993		
DOCUMENT TYPE:	Utility		
FILE SEGMENT:	Granted		
PRIMARY EXAMINER:	Feisee, Lila		
ASSISTANT EXAMINER:	Kaufman, Claire M.		
LEGAL REPRESENTATIVE:	Sterne, Kessler, Goldstein & Fox P.L.L.C.		
NUMBER OF CLAIMS:	46		
EXEMPLARY CLAIM:	22		
NUMBER OF DRAWINGS:	55 Drawing Figure(s); 47 Drawing Page(s)		
LINE COUNT:	3020		

CAS INDEXING IS AVAILABLE FOR THIS PATENT.

AB A DNA enhancer element and the use of this syndecan enhancer element to regulate the expression of genes are provided. DRWD . . . produce a supershift with labelled motif 3 and nuclear extracts deemed from FGF-2 treated 3T3 NIH cells. A gel retardation gel as shown in FIG. 1SA was run and exposed to UV light. The specific bands, representing the bound protein-DNA complex, were cut out, eluted overnight, and loaded onto an SDS-PAGE gel to analyze their molecular weight. Two reproducible bands for the motif 3 binding protein are shown. The molecular weight of the nuclear factors were approximated

by subtracting the calculated molecular weight of each oligonucleotide from the complex molecular weight.

DETD The FIN-1 protein has been isolated and has a molecular weight of 50 kDa as determined by SDS-PAGE.

DETD SDS-PAGE and Western Blot--For western blot experiments, cells were cultured 24 hours with or without growth factor(s). Syndecan ectodomain containing material released from the cell surface by trypsin treatment was fractionated on SDS-PAGE gradient (2-15%) gel (O'Farrel, J. Biol. Chem. 250:4007-4021 (1975)). After electrophoresis, samples were transferred onto a Zeta-Probe membrane by electroblotting with a 2005 Transphor apparatus (LKB). The syndecan antigen on the filter was detected with radioiodinated mAB 281-2 and the filter was washed, as described above for slot blot analysis.

DETD . . . were size-separated on a 1% agarose formaldehyde gel, transferred to a GeneScreen Plus.TM. membrane (New England Nuclear) and hybridized with a multi-prime (Amersham) labeled partial cDNA clone for mouse syndecan (PM4) (Saunders et al., J. Cell Biol. 108:1547-1556 (1989)). After hybridization, the membrane was washed in 2+ SSC and 1.0% SDS at 65° C. (high stringency conditions). For rehybridization with glyceraldehyde-3-phosphate-dehydrogenase (GAPDH; Fort et al., Nucleic Acid Res. 13:1431-1442 (1985)), the bound PM-4 probe was removed as recommended by the manufacturer of the filter (NEN).

DETD . . . run, it was exposed to 245 nm UV-light (3600J/em.sup.2) in a Strategene crosslinker. The gel was exposed for several hours, the specific bands were cut out, eluted overnight at 4° C., precipitated with ethanol, resuspended in Laemmli buffer, denatured at 95° C. for 5 minutes, and loaded onto a 10% SDS-PAGE together with a .sup.14 C-labeled molecular weight markers to analyze their molecular weights. The SDS-PAGE gel is shown in FIG. 15C, with the position of the molecular weight markers shown at the left. Lanes 1-5 correspond to motifs 1-5, respectively. The molecular weights of the nuclear factors were estimated after subtracting the mass of the oligonucleotide from the complex mass as indicated below:

DETD This experiment shows a reproducible 46 kDa band for motif 1 and two bands, 78 kDa and 50 kDa, for motif 3. These values have a margin of error of about ±3 kDa.

L83 ANSWER 3 OF 9 PCTFULL COPYRIGHT 2011 LNU on STN

ACCESSION NUMBER:	2011034605 PCTFULL Full-text
ENTRY DATE:	20110328
UPDATE DATE:	20110613
ENTRY DATE (FULLTEXT):	20110328
DATA ENTRY DATE:	20110324
DATA UPDATE DATE:	20110524
TITLE (ENGLISH):	COILED COIL AND/OR TETHER CONTAINING PROTEIN COMPLEXES AND USES THEREOF
TITLE (FRENCH):	COMPLEXES PROTEIQUES CONTENANT UNE SUPER-HELICE ET/OU UNE ATTACHE ET LEURS UTILISATIONS
INVENTOR(S):	CHRISTENSEN, Erin H., c/o Genentech, Inc., 1 DNA Way, South San Francisco, California 94080, US, [NAT: US, RES: US], for US only EATON, Dan L., c/o Genentech, Inc., 1 DNA Way, South San Francisco, California 94080, US, [NAT: US, RES: US], for US only VENDEL, Andrew C., c/o Genentech, Inc., 1 DNA Way, South San Francisco, California 94080, US, [NAT: US, RES: US], for US only WRANIK, Bernd, c/o Genentech, Inc., 1 DNA Way, South San Francisco, California 94080, US, [NAT: DE, RES:

US], for US only
PATENT APPLICANT(S): GENENTECH, INC., 1 DNA Way, South San Francisco, California 94080, US, [NAT: US, RES: US], for designated states AE AG AM AO AU AZ BA BB BF BH BJ BR BW BY BZ CA CF CG CI CL CM CO CR CU DM DO DZ EC EG GA GD GE GH GM GN GQ GT GW HN ID IL JP KE KG KM KN KP KR KZ LA LC LK LR LS LY MA MD ME MG ML MN MR MW MX MY MZ NA NE NG NI NZ OM PE PG PH RS RU SC SD SG SL SN ST SV SY SZ TD TG TH TJ TM TN TT TZ UA UG UZ VC VN ZA ZM ZW
F. HOFFMANN-LA ROCHE AG, Grenzacherstrasse 124, CH-4070 Basel, CH, [NAT: CH, RES: CH], for designated states AL AT BE BG CH CN CY CZ DE DK EE ES FI FR GB GR HR HU IE IN IS IT LT LU LV MC MK MT NL NO PL PT RO SE SI SK SM TR
CHRISTENSEN, Erin H., c/o Genentech, Inc., 1 DNA Way, South San Francisco, California 94080, US, [NAT: US, RES: US], for US only
EATON, Dan L., c/o Genentech, Inc., 1 DNA Way, South San Francisco, California 94080, US, [NAT: US, RES: US], for US only
VENDEL, Andrew C., c/o Genentech, Inc., 1 DNA Way, South San Francisco, California 94080, US, [NAT: US, RES: US], for US only
WRANIK, Bernd, c/o Genentech, Inc., 1 DNA Way, South San Francisco, California 94080, US, [NAT: DE, RES: US], for US only
AGENT: SHIN, Elinor K. et al., Genentech, Inc., 1 DNA Way, MS 49, South San Francisco, California 94080, US
LANGUAGE OF FILING: English
LANGUAGE OF PUBL.: English
DOCUMENT TYPE: Patent; (Fulltext)
PATENT INFORMATION: WO 2011034605 A2 20110324
DESIGNATED STATES:
W: AE AG AL AM AO AT AU AZ BA BB BG BH BR BW BY BZ CA CH CL CN CO CR CU CZ DE DK DM DO DZ EC EE EG ES FI GB GD GE GH GM GT HN HR HU ID IL IN IS JP KE KG KM KN KP KR KZ LA LC LK LR LS LT LU LY MA MD ME MG MK MN MW MX MY MZ NA NG NI NO NZ OM PE PG PH PL PT RO RS RU SC SD SE SG SK SL SM ST SV SY TH TJ TM TN TR TT TZ UA UG US UZ VC VN ZA ZM ZW
RW (ARIPO): BW GH GM KE LR LS MW MZ NA SD SL SZ TZ UG ZM ZW
RW (EAPO): AM AZ BY KG KZ MD RU TJ TM
RW (EPO): AL AT BE BG CH CY CZ DE DK EE ES FI FR GB GR HR HU IE IS IT LT LU LV MC MK MT NL NO PL PT RO SE SI SK SM TR
RW (OAPI): BF BJ CF CG CI CM GA GN GQ GW ML MR NE SN TD TG
APPLICATION INFO.: WO 2010-US2546 20100916
PRIORITY INFO.: US 2009-243105P 20090916
US 2009-266992P 20091204

ABEN

The invention provides engineered protein complexes constructed using a coiled coil and/or a tether and methods for making, using, and purifying such complexes, such as multispecific antibodies or other multispecific Fc containing complexes.

ABFR

La presente invention concerne des complexes proteiques genetiquement modifies construits a l'aide d'une helice et/ou d'une attache et des procedes pour produire, utiliser et purifier de tels complexes tels, que des anticorps multispecifiques ou d'autres complexes multispecifiques contenant Fc.

DETDEN . . .
invention features a method of maintaining a coiled coil containing antibody in solution. This method comprises maintaining the antibody in the presence of a chaotropic agent or mild detergent. Examples, of chaotropic agents or mild detergents that may be used in this method include Arginine, Guanidine-HCl, urea, lithium perchlorate, Histidine, Sodium Dodecyl Sulfate (SDS), Tween, Triton, and NP-40.

DETDEN
(MW=50528 and 50767) are within the margin of error of the experimentally observed masses indicated in the graph of the mass spectrometry results for the respective construct.

DETDEN . . .
are a series of graphs of mass spectrometry results and schematic diagrams showing that the coiled coil can be cleaved from an exemplary one-armed a-EGFR antibody using Lys-C endopeptidase. The theoretical masses of the one-armed antibody with a coiled coil (MW=1091 12), and the one-armed antibody without a coiled coil (MW=100419) are within the margin of error of the experimentally observed masses indicated in the graph of the mass spectrometry results for the respective construct.

DETDEN . . .
than 95% by weight of protein as determined by the Lowry method, and most preferably more than 99% by weight, (2) to a degree sufficient to obtain at least 15 residues of N-terminal or internal amino acid sequence by use of a spinning cup sequenator, or (3) to homogeneity by SDS-PAGE under reducing or

DETDEN . . .
and can include enzymes, hormones, and other proteinaceous or nonproteinaceous solutes. In preferred embodiments, the antibody will be purified (1) to a degree sufficient to obtain at least 15 residues of N-terminal or internal amino acid sequence by use of a spinning cup sequenator, or (2) to homogeneity by SDS-PAGE under non-reducing or reducing conditions using Coomassie blue or, preferably, silver stain. Isolated antibody includes antibodies in situ within recombinant cells, because at least one component of the polypeptide natural environment will not be present. Ordinarily, however, isolated polypeptide will be prepared by at least one purification step. By "linked". . .

DETDEN
phenoxylpolyethoxylethanol), Nonidet P-40 (octyl phenoxylpolyethoxylethanol), and Sodium Dodecyl Sulfate (SDS).

DETDEN
Standard protein purification methods known in the art can be employed. The following procedures are exemplary of suitable purification procedures: fractionation on immunoaffinity or ion-exchange columns, ethanol precipitation, reverse phase HPLC, chromatography on silica or on a cation-exchange resin such as DEAE, chromatofocusing, SDS-PAGE, ammonium sulfate precipitation, and gel filtration using, for example, Sephadex G-75.

DETDEN . . .
to remove contaminants non-specifically bound to the solid phase. The antibody of interest may be recovered from the solid phase by elution into a solution containing a chaotropic agent or mild detergent. Exemplary chaotropic agents and mild detergents include, but are not limited to,

Guanidine-HCl, urea, lithium perclorate, Arginine, Histidine, SDS (sodium dodecyl sulfate), Tween, Triton, and NP-40, all of which are commercially available. Diluting the antibody into a solution containing a chaotropic agent or mild detergent after elution from the column (e.g., mAbSure column) maintains the stability of the antibody post elution and allows for the efficient removal of the coiled coil. . .

DETDEN . . .
Bakerbond ABX®resin (J. T. Baker, Phillipsburg, NJ) is useful for purification. Other techniques for protein purification such as fractionation on an ion-exchange column, ethanol precipitation, Reverse Phase HPLC, chromatography on silica, chromatography on heparin SEPHAROSE® chromatography on an anion or cation exchange resin (such as a polyaspartic acid column), chromatofocusing, SDS-PAGE, and ammonium sulfate

DETDEN
In one embodiment, the antibody of interest is recovered from the solid phase of a column by elution into a solution containing a chaotropic agent or mild detergent. Exemplary chaotropic agents and mild detergents include, but are not limited to, Guanidine-HCl, urea, lithium perclorate, Arginine, Histidine, SDS (sodium dodecyl sulfate), Tween, Triton, and NP-40, all of which are commercially available.

DETDEN . . .
with a secondary wash buffer (50 mM phosphate; 300 mM NaCl; 10% glycerol pH 6.0), which elutes nonspecifically bound protein. After reaching A280 baseline again, the column is developed with a 0 to 500 mM Imidazole gradient in the secondary wash buffer. One mL fractions are collected and analyzed by SDS- PAGE and silver staining or Western blot with Nisup2+-NTA-conjugated to alkaline phosphatase (Qiagen). Fractions containing the eluted Hisi0-tagged antibody are pooled and dialyzed against loading buffer.

DETDEN
chromatography. The antibody of interest may be recovered from the solid phase of the column by elution into a solution containing a chaotropic agent or mild detergent. Exemplary chaotropic agents and mild detergents include, but are not limited to, Guanidine-HCl, urea, lithium perclorate, Arginine, Histidine, SDS (sodium dodecyl sulfate), Tween, Triton, and NP-40, all of which are commercially available. c. Optimized purification technique

DETDEN
In addition to Arginine, other chaotropic agents or mild detergents that can be used in the above purification protocol after the initial Protein A column step include, but are not limited to, Guanidine-HCl, urea, lithium perclorate, Histidine, SDS (sodium dodecyl sulfate), Tween, Triton, and NP-40, all of which are commercially available. Diluting the antibody into a solution containing a chaotropic agent or mild detergent after elution from the initial Protein A containing column (e.g., mAbSure column) maintains the stability of the antibody post elution and allows for the efficient removal. . .

DETDEN
In addition to Arginine, other chaotropic agents or mild detergents that can be used in the above purification protocol after the initial mAbSure resin column step include, but are not limited to, Guanidine-HCl, urea, lithium perclorate, Histidine, SDS (sodium dodecyl sulfate), Tween, Triton, and NP-40, all of which are commercially available. Diluting the antibody into a solution containing a chaotropic agent or mild

detergent after elution from the initial Protein A containing column (e.g., mAbSure column) maintains the stability of the antibody post elution and allows for the efficient removal. . .

DETDEN . . .
functional properties of exemplary engineered antibodies were also characterized biochemically. EGFR-expressing NR6 cells were plated in 12-well plates. Following serum starvation cells were pre-incubated with various concentrations of antibodies for 2 hours at 37°C. Subsequently, cells were stimulated with the TGFa for 12 minutes. Whole cell lysates were subjected to SDS-PAGE analysis, and immunoblots were probed with anti- phosphotyrosine, anti-phosphoAkt, or anti-tubulin as a loading control (Figure 24). These results show that the exemplary a-EGFR(D1.5)/Anti-HER2 (antibody 1) engineered antibody, like the D 1.5 IgG1 control antibody, inhibited TGFa-induced phosphorylation in EGFR- expressing NR6 cells in a dose-dependent manner.

CLMEN
55. The method of claim 53 or 54, wherein said chaotropic agent or mild detergent is Arginine, Guanidine-HCl, urea, lithium perchlorate, Histidine, Sodium Dodecyl Sulfate (SDS), Tween, Triton, or NP-40.

L83 ANSWER 4 OF 9 PCTFULL COPYRIGHT 2011 LNU on STN

ACCESSION NUMBER:	2001083534 PCTFULL Full-text
ENTRY DATE:	20101209
UPDATE DATE:	20101209
ENTRY DATE (FULLTEXT):	20101209
DATA UPDATE DATE:	20080627
TITLE (ENGLISH):	ANTI-FREEZE PROTEINS, THEIR PRODUCTION AND USE
TITLE (FRENCH):	PROTEINES ANTIREFRIGERANTES, PRODUCTION ET UTILISATION DE CELLES-CI
INVENTOR(S):	BERRY, Mark, John, Unilever Research Colworth, Colworth House, Sharnbrook, Bedford, Bedfordshire MK44 1LQ, GB DOUCET, Charlotte, Juliette, c/o University of York, Department of Boilogy, The Plant Laboratory, Heslington, Yorkshire YO1 5YM, GB LUNDHEIM, Rolv, Sigmund, Queens Maud College, Thonnig Owesens GT, 18, N-N-7044 Trondheim, NO SEVILLA, Marie-Pierre, Unilever Research Colworth, Colworth House, Sharnbrook, Bedford, Bedfordshire MK44 1LQ, GB WHITEMAN, Sally-Anne, Unilever Research Colworth, Colworth House, Sharnbrook, Bedford, Bedfordshire MK44 1LQ, GB
PATENT APPLICANT(S):	UNILEVER PLC, Unilever House, Blackfriars, London EC4P 4BQ, GB UNILEVER NV, Weena 455, NL-3013 AL Rotterdam, NL HINDUSTAN LEVER LIMITED, Hindustan Lever House, 165/166 Backbay Reclamation, Maharashtra, 400 020 Mumbai, IN
AGENT:	EVANS, Jacqueline, Gail, Victoria, Unilever PLC, Patent Department, Colworth House, Sharnbrook, Bedford, Bedfordshire MK44 1LQ, GB
LANGUAGE OF FILING:	English
LANGUAGE OF PUBL.:	English
DOCUMENT TYPE:	Patent; (Fulltext)
PATENT INFORMATION:	WO 2001083534 A1 20011108
DESIGNATED STATES:	
W:	AE AG AL AM AT AU AZ BA BB BG BR BY BZ CA CH CN CR CU CZ DE DK DM DZ EE ES FI GB GD GE GH GM HR HU ID IL IN

IS JP KE KG KP KR KZ LC LK LR LS LT LU LV MA MD MG MK
MN MW MX MZ NO NZ PL PT RO RU SD SE SG SI SK SL TJ TM
TR TT TZ UA UG US UZ VN YU ZA ZW
RW (ARIPO): GH GM KE LS MW MZ SD SL SZ TZ UG ZW
RW (EAPO): AM AZ BY KG KZ MD RU TJ TM
RW (EPO): AT BE CH CY DE DK ES FI FR GB GR IE IT LU MC NL PT SE
TR
RW (OAPI): BF BJ CF CG CI CM GA GN GW ML MR NE SN TD TG
APPLICATION INFO.: WO 2001-EP3927 20010406
PRIORITY INFO.: GB 2000-10314 20000427

ABEN

Antifreeze proteins which can be derived from the lichen Nephroma arcticum and proteins having antifreeze activity having an amino acid sequence part of which shows at least 80% overlap with the amino acid sequence L-V-I-G-S-T-A-Q(E)-N-F-G-V-V(S)-A-A-A-T, as well as modified versions thereof. Methods for their preparation, their use in food processing and food compositions comprising them are also described.

ABFR

L'invention concerne des proteines antirefrigerantes pouvant etre derivees du lichen Nephroma arcticum et des proteines dotees d'une activite antirefrigerante possedant une sequence aminoacide dont une partie presente au moins 80 % de chevauchement avec la sequence aminoacide L-V-I-G-S-T-A-Q(E)-N-F-G-V-V(S)-A-A-A-T, ainsi que des versions modifiees de celle-ci. L'invention concerne egalement des procedes de fabrication de ces proteines et d'utilisation de ces dernieres dans le traitement alimentaire ainsi que des compositions alimentaires comportant lesdites proteines.

DETDEN . . .

major antifreeze protein has so far been identified by the inventors and its sequence has been partly determined. The invention also encompasses other proteins that may be contributory to the antifreeze activity in this lichen species.The major AFP isolated from Nephroma arcticum has an apparent molecular weight, as judged by SDS-polyacrylamide gel electrophoresis, of around 29 kDa, (although given the limitations of the technique there is a likely margin of error of +/-4 kDa on this value). The N-terminal amino acid sequence of this proteins has been determined to be: L-V-I-G-S-T-A-Q (E)-N-F-G-V-V (S)-A-A-A-T There appears to be some sequence heterogeneity at positions 8 (major form Q with E as a minor variant) and 13 (major form V with S as a minor variant) as. . .

DETDEN . . .

in buffer B (50 mM Tris/HC1 pH 7.5). The flow rate was 40 gl/min and 50 pl fractions were collected. The active fractions after the gel filtration step were pooled and concentrated and used for N-terminal analysis as described in example 10.Conclusion During the purification protocol given above, gel electrophoresis (SDS-PAGE with silver staining) was used to identify the AFP. This technique consistently identified a negatively staining band at 29kDa that co-purified with AFP activity (as adjudged by the Splat assay). Therefore, this protein was known to be the AFP and it was this band that was N-terminal sequenced as described in example 10.EXAMPLE 10 Determination of the N-terminal sequence of the AFP derived from Nephroma arcticum.90 pLI purified N. arcticum AFP sample (prepared as described in example 9) was applied equally to four adjacent lanes and separated by SDS-PAGE prior to western blotting onto PVDF membrane. The membrane had been soaked in methanol and the blotting buffer used was 10 mM CAPS, pH11 plus 10 % methanol. The membrane was stained with Ponceau stain and the relevant bands marked with a needle before removal of the stain with water..

. .

L83 ANSWER 5 OF 9 PCTFULL COPYRIGHT 2011 LNU on STN
ACCESSION NUMBER: 1998024921 PCTFULL Full-text
ENTRY DATE: 20101211
UPDATE DATE: 20110502
ENTRY DATE (FULLTEXT): 20101211
DATA UPDATE DATE: 20110427
TITLE (ENGLISH): SYNDECAN ENHANCER ELEMENT AND ITS USE FOR TARGETING GENE EXPRESSION
TITLE (FRENCH): ELEMENT STIMULATEUR DE SYNDECANE ET SON UTILISATION POUR CIBLER L'EXPRESSION GENIQUE
INVENTOR(S): JALKANEN, Markku, Rauvolantie 79, FIN-20760 Piispanristi, FI
JAAKKOLA, Panu, Kellonsoittajankatu 13 B 20, FIN-20500 Turku, FI
VIHINEN, Tapani, Kaskenkatu 11 C 54, FIN-20700 Turku, FI
PATENT APPLICANT(S): OY BIOTIE THERAPIES, LTD., BioCity, Tykistoenkatu 6, FIN-20520 Turku, FI
AGENT: ORION CORPORATION, Orion Pharma, Industrial Property Rights, P.O. Box 65, FIN-02101 Espoo, FI
LANGUAGE OF FILING: English
LANGUAGE OF PUBL.: English
DOCUMENT TYPE: Patent; (Fulltext)
PATENT INFORMATION: WO 9824921 A1 19980611
DESIGNATED STATES:
W: AL AM AU AZ BA BG BR BY CA CN CZ EE GE HU ID IL IS JP KG KR KZ LT LV MD MK MX NO NZ PL RO RU SG SI SK TJ TM TR UA US UZ YU
RW (EAPO): AM AZ BY KG KZ MD RU TJ TM
RW (EPO): AT BE CH DE DK ES FI FR GB GR IE IT LU MC NL PT SE
APPLICATION INFO.: WO 1997-FI748 19971202
PRIORITY INFO.: US 1996-8760534 19961202

ABEN
A syndecan enhancer element, novel proteins that activate the enhancer element, non-human transgenic animals comprising this enhancer element linked to a structural gene, and the use of this enhancer element to regulate the expression of syndecan and other genes are also provided. The enhancer element can also be used to target expression of a gene to wound sites.

ABFR
L'invention concerne un element stimulateur de syndecane, des nouvelles proteines qui activent ledit element activateur, des animaux transgeniques non humains comprenant ledit element stimulateur lie a un gene structural et l'utilisation de cet element stimulateur pour la regulation de l'expression de syndecane et d'autres genes. Ledit element stimulateur peut egalement etre utilise pour diriger l'expression d'un gene sur des sites de blessures.

DETDEN
The FIN-1 protein has been isolated and has a molecular weight of 50 kDa as determined by SDS-PAGE.

DETDEN . . .
gel was run, it was exposed to 245 nm UV-light (3600J/em2) in a Strategene crosslinker. The gel was exposed for several hours, the specific bands were cut out, eluted overnight at 4EC, precipitated with ethanol, resuspended in

Laemmli buffer, denatured at 95EC for 5 minutes, and loaded onto a 10% SDS-PAGE together with a 14Clabeled molecular weight markers to analyze their molecular weights. TheSDS-PAGE gel is shown in Figure <RTI ID=21.4>1 it,</RTI> with the position of the molecular weight markers shown at the left.STDC0574 Lanes 1-5 correspond to motifs 1-5, respectively. The molecular weights of the nuclear factors were estimated after. . . mass as indicated below: Motif MW Oligo +Factor MW Oligo MW Factor <RTI ID=22.1>66</RTI> kDa 20 kDa 46 kDa 3 <RTI ID=22.2>62 kDa; 90 kDa</RTI> 12 kDa <RTI ID=22.3>50 kDa; 78 kDa</RTI> This experiment shows a reproducible 46 kDa band for motif 1 and two bands, 78 kDa and 50 kDa, for motif 3. These values have a margin of error of about <RTI ID=22.4>+3</RTI> kDa.

L83 ANSWER 6 OF 9 PCTFULL COPYRIGHT 2011 LNU on STN
ACCESSION NUMBER: 1994023067 PCTFULL Full-text
ENTRY DATE: 20101213
UPDATE DATE: 20101213
ENTRY DATE (FULLTEXT): 20101213
DATA UPDATE DATE: 20080224
TITLE (ENGLISH): TUMOR-ASSOCIATED ANTIGENS RECOGNIZED BY T CELLS AND THE USES OF THESE ANTIGENS
TITLE (FRENCH): ANTIGENES ASSOCIES A DES TUMEURS RECONNUS PAR LES LYMPHOCYTES ET UTILISATIONS DE CES ANTIGENES
INVENTOR(S): REILLY, Edward, B.
EISEN, Herman, N.
TSOMIDES, Theodore
PATENT APPLICANT(S): ABBOTT LABORATORIES
LANGUAGE OF FILING: English
LANGUAGE OF PUBL.: English
DOCUMENT TYPE: Patent; (Fulltext)
PATENT INFORMATION: WO 9423067 A1 19941013
DESIGNATED STATES:
W: AU CA JP KR
RW (EPO): AT BE CH DE DK ES FR GB GR IE IT LU MC NL PT SE
APPLICATION INFO.: WO 1994-US3507 19940331
PRIORITY INFO.: US 1993-8040800 19930331

ABEN

This invention relates to the field of tumor immunology, and specifically to a novel family of melanoma-specific antigens recognized by T cells. These antigens, like all T cell epitopes, are in the form of small peptides associated with major histocompatibility complex antigens on the cell surface. Methods and materials for purification and sequence determination of these peptides are presented. Also presented are applications for their use in cancer diagnostics and therapy.

ABFR

Cette invention concerne le domaine de l'immunologie des tumeurs et plus particulierement une nouvelle famille d'antigenes specifiques aux melanomes, reconnus par les lymphocytes T. Ces antigenes, comme tous les epitopes des lymphocytes T se presentent sous la forme de petits peptides associes a des antigenes du complexe majeur d'histocompatibilite sur la surface des cellules. L'invention concerne egalement des procedes et des produits pour la purification et la determination en sequences de ces peptides. Sont egalement presentees des applications dans le domaine du diagnostic et de la therapie du cancer.

DETDEN . . .

to 20 amino acids in length, more preferably between 5-15, and most preferably between 7 to 1 2 amino acids in length. Examples of the T cell-specific melanoma antigens are peptides such as mel Ag 906 or mel Ag 1007. The molecular weight of mel Ag 906 is about 906 Dalton (D) with a + 10% margin of error. The molecular weight of mel Ag 1007 is about 1007 Dalton (D) with a + 10% margin of error.

DETDEN . . .
to 20 amino acids in length, more preferably between 5-15, and most preferably between 7 to 1 2 amino acids in length. Examples of the T cell-specific melanoma antigens are peptides such as mel Ag 906 or mel Ag 1007. The molecular weight of mel Ag 906 is about 906 Dalton (D) with a + 10% margin of error. The molecular weight of mel Ag 1007 is about 1007 Dalton (D) with a + 10% margin of error.

DETDEN
Figure 3 shows the results of a typical elution profile from the mouse immunoglobulin and the two successive PA2.1 columns. Figure 3 presents the HLA-A2 affinity purification from 660 mel cells. HLA purity was assessed by SDS/PAGE. Yields and purity were further determined by quantitative amino acid analysis.

DETDEN . . .
and molecular weight similar to the method disclosed in Hunt, D. F., et al. (1992) Science 255: 1261. There were several peptides in the fractions. The two most prevalent peptides, designated mel Ag 906 and mel Ag 1007 were identified. The molecular weight of mel Ag 906 is about 906 Dalton (D) with a + 10% margin of error. The molecular weight of mel Ag 1007 is about 1007 Dalton (D) with a + 10% margin of error. The amino acid sequence can be determined by similar tandem mass spectrometry.

L83 ANSWER 7 OF 9 EPFULL COPYRIGHT 2011 EPO/FIZ KA/LNU on STN

ACCESSION NUMBER:	2001:50001 EPFULL Full-text
UPDATE DATE PUBLICAT.:	20070516
DATA UPDATE DATE:	20070516
DATA UPDATE WEEK:	200720
TITLE (ENGLISH):	ANTI-FREEZE PROTEINS, THEIR PRODUCTION AND USE
TITLE (FRENCH):	PROTEINES ANTIREFRIGERANTES, PRODUCTION ET UTILISATION DE CELLES-CI
TITLE (GERMAN):	ANTI-GEFRIER PROTEINE, DEREN HERSTELLUNG UND VERWENDUNG
INVENTOR(S):	BERRY, Mark John,Unilever Research Colworth, Colworth House,Sharnbrook, Bedford, Bedfordshire MK44 1LQ, GB; DOUCET, Charlotte Juliette,University of York, Department of Biology,The Plant Laboratory, Heslington,Yorkshire YO1 5YM, GB; LUNDHEIM, Rolv Sigmund, Queens Maud College, Thonnig Owesens GT,18,N-N-7044 Trondheim, NO; SEVILLA, Marie-Pierre, 5 rue Jacques PrevertBP 33, 31520 Ramonville saint agne, FR; WHITEMAN, Sally-Anne,Unilever Research Colworth, Colworth House,Sharnbrook, Bedford,Bedfordshire MK44 1LQ, GB
PATENT APPLICANT(S):	UNILEVER PLC, Unilever House, Blackfriars, London EC4P 4BQ, GB; UNILEVER N.V., Weena 455, 3013 AL Rotterdam, NL
PATENT APPL. NUMBER:	200923; 200916
PA DESIGNATED STATES:	CY GB IE; AT BE CH DE DK ES FI FR GR IT LI LU MC NL PT

	SE TR
AGENT:	Hugot, Alain, et al, Unilever Patent Group, Colworth HouseSharnbrookBedford, MK44 1LQ, GB
AGENT NUMBER:	61541
DOCUMENT TYPE:	Patent
LANGUAGE OF FILING:	English
LANGUAGE OF PUBL.:	English
LANGUAGE OF PROCEDURE:	English
LANGUAGE OF TITLE:	German; English; French
PATENT INFO TYPE:	EPB1 Granted patent
PATENT INFORMATION:	
PATENT INFORMATION:	

NUMBER	KIND	DATE
NUMBER	KIND	DATE
EP 1276763	B1	20040225
WO 2001083534		20011108

DESIGNATED STATES:	AT BE CH CY DE DK ES FI FR GB GR IE IT LI LU MC NL PT SE TR
APPLICATION INFO.:	EP 2001-919437 A 20010406
	WO 2001-EP3927 A 20010406
PRIORITY INFO.:	GB 2000-10314 A 20000427
CITED PATENT LIT.:	WO 9804148 A (INID56)
	WO 9937673 A (INID56)

DETDEN

[0023] The major AFP isolated from Nephroma arcticum has an apparent molecular weight, as judged by SDS-polyacrylamide gel electrophoresis, of around 29 kDa, (although given the limitations of the technique there is a likely margin of error of +/- 4 kDa on this value). The N-terminal amino acid sequence of this proteins has been determined to be:
L-V-I-G-S-T-A-Q(E)-N-F-G-V-V(S)-A-A-A-T

DETDEN

[0094] During the purification protocol given above, gel electrophoresis (SDS-PAGE with silver staining) was used to identify the AFP. This technique consistently identified a negatively staining band at 29kDa that co-purified with AFP activity (as adjudged by the Splat assay). Therefore, this protein was known to be the AFP and it was this band that was N-terminal sequenced as described. . .

DETDEN

[0095] 90 µl purified N. arcticum AFP sample (prepared as described in example 9) was applied equally to four adjacent lanes and separated by SDS-PAGE prior to western blotting onto PVDF membrane. The membrane had been soaked in methanol and the blotting buffer used was 10 mM CAPS, pH11 plus 10 % methanol. The membrane was stained with Ponceau stain and the relevant bands marked with a needle before removal of the stain with water.. . .

L83 ANSWER 8 OF 9 FRFULL COPYRIGHT 2011 LNU on STN

ACCESSION NUMBER:	2758144 FRFULL ED 20100221 Full-text UP 20101124
TITLE (ENGLISH):	POLYNUCLEOTIDE CODING FOR A POLYPEPTIDE OF 27 KD OF MYCOBACTERIES PERTAINING TO THE COMPLEX OF MYCOBACTERIUM TUBERCULOSIS, APPLICATION TO THE DIAGNOSIS AND THE PREVENTION OF TUBERCULOSIS

TITLE (FRENCH): POLYNUCLEOTIDE CODANT POUR UN POLYPEPTIDE DE 27 KD DE MYCOBACTERIES APPARTENANT AU COMPLEXE DE MYCOBACTERIUM TUBERCULOSIS, APPLICATION AU DIAGNOSTIC ET A LA PREVENTION DE LA TUBERCULOSE
INVENTOR(S): GUESDON JEAN LUC; CHEVRIER DANIELE
PATENT APPLICANT(S): INSTITUT PASTEUR
PATENT APPL. COUNTRY: FR
LANGUAGE OF FILING: French
LANGUAGE OF PUBL.: French
DOCUMENT TYPE: Patent
PATENT INFO TYPE: FRB1 PATENT OF INVENTION (SECOND PUBLICATION) (FROM 2,000,000)
PATENT INFORMATION:

	NUMBER	KIND	DATE
	FR 2758144	B1	19990402
APPLICATION INFO.:	FR 1997-100	A	19970108
PRIORITY INFO.:	FR 1997-100	A	19970108 *

DETDEN . . . the expression of the sequence codanle onl identified upstream el downstream from the latter.Thus, the invention relates to a polynucleotide of 2805 pairs of bases, specific of the complex of tuberculosis. This polynucleotide inclui the sequence corresponding to a gene of structure called p27, which codes for a protein of molecular weight ofapproximately 27 kDa, appreciated with a margin of error of 10%.By gene of structure for purposes of this invention, one understands a polynucleotide coding for a protein, a polypeptide or afragment of the latter, the aforementioned polynucleotide not understanding that the sequence corresponding to the open framework ofreading (ORF), which excludes the sequences on the side 5' of the. . . ADN.On a purely illustrative basis, conditions of stringence of the stage of hybridization for purposes of defining the fragments polynucleotidic described above, are advantageously following hybridization is carried out at a preferential temperature of 65°C, inthe presence of 5 plug 6 X SSC, 5 X of solution of Denhardt, 0,5% SDS and 100 ug/ml of ADN of salmon sperm. X SSC corresponds to 0,15 M NaCl and 0,05M citrate of Na and a solution of 1 X Denhardt corresponds to 0,02% Ficoll, polyvinylpyrrolidone 0,02% and 0,02% of serum bovine albumin. the 10 stages of washing can, for example,being the following ones:-two washings of 5 min, preferentially with 65°C, in a plug 2 X SSC and 0,1% SDS;-a washing of 30 min, preferentially with 65°C, in a plug 2 X SSC and 0,1% SDS;-a washing of 10 min, preferentially with 65°C, in a plug of 1 X SSC and 0,1% SDS .The invention also relates to a polynucleotide including/understanding the open framework of reading coding for a polypeptide of a molecular weight of about 27 kD. According to aof the aforesaid mode of realization preferred polynucleotide, it consists of a sequence presenting an open framework of reading (ORF) which comprises at its. . .

. . .

the stage of hybridization for purposes specifically of detecting atarget ADN of a mycobactery belonging to the complex of mycobacterium tuberculosis, can advantageously be as follows: hybridization iscarried out at a preferential temperature of 65°C, in the presence of plug 6 X SSC, 5 X of solution of Denhardt, 0,5% SDS and 100 ug/ml of ADN of sperm of 15 salmon. X SSC corresponds to 0,15 M NaCl and 0,05M citrate of Na and a solution of 1 X Denhardt corresponds to 0,02% Ficoll, 0,02% of plyvinylpyrrolidone and 0,02% of serum bovine albumin.The stages of washing can, for example, being the following

ones: 20-two washings of 5 min, preferentially with 65°C, in a plug 2 X SSC and 0,1% SDS;-a washing of 30 min, preferentially with 65°C, in a plug 2 X SSC and 0,1% SDS;-a washing of 10 min, preferentially with 65°C, in a plug of 0,1 X SSC and 0,1% SDS.The not marked sequences can be used directly as probes, however the sequences are generally marked by a radioactive element (32P, 35S, 3H, I2I) or by a not-radioactive molecule (biotine, acetylaminofluorene, digoxigenine, 5-bromo-desoxyuridine, fluorescein) to obtain probes usable for many applications.Examples of nonradioactive markings of probes are described, for example, in. . .

DETDFR I0 A titre illustratif, des conditions de stringence de l'etape d'hybridation aux fins de definir les fragments polynucleotidiques decrits ci-dessus, sont avantageusenlcnt les suivantes l'hybridation est realisee a une temperature preferentielle de 65°C, en presence de tampon 6 x SSC, 5 x de solution de Denhardt, 0,5% SDS et 100 tg/ml d'ADN de spernle de saumon.

- deux lavages de 5 rein, preferentiellement a 65°C, dans un tampon 2 x SSC et 0,1% SDS; - un lavage de 30 rein, preferentiellement a 65°C, dans un tampon 2 x SSC et 0,1% SDS; - un lavage de 10 rein, preferentiellement a 65°C, dans un tampon de I x SSC et 0,1% SDS.

['hybridation est realisee a une temperature preferentielle de 65°C, en presence de tampon 6 x SSC, 5 x de solution de Denhardt, 0,5% SDS et 100 p.g/ml d'ADN de sperme de saumon.

- deux lavages de 5 min, preferentieilement a 65°C, dans un tampon 2 x SSC et 0,1% SDS; - un lavage de 30 min, preferentiellement a 65°C, dans un tampon 2 x SSC et 0,1% SDS; - un lavage de 10 min, preferentieilement a 65°C, dans un tampon de 0,1 x SSC et 0,1% SDS.

L'ADN est extrait par remise en suspension du culot avec 50 bll de NaOH 0,1 M contenant du NaCI 2 M et du SDS 0,5%. Le melange est incube a 95°C pendant 15 minutes, au melange reactionnel on ajoute 400 pl de Tris-HC1 0,1M pli 7. L'ADN est extrait 3 lois at.

L83 ANSWER 9 OF 9 FRFULL COPYRIGHT 2011 LNU on STN

ACCESSION NUMBER:	2681076 FRFULL	ED 20100221 EDTX 20040305 Full-text
		UP 20100929
TITLE (ENGLISH):	RECOMBINING DNA CODING FOR A PROTEIN HAS ACTIVITY ENDOCHITINASE.	
TITLE (FRENCH):	ADN RECOMBINANT CODANT POUR UNE PROTEINE A ACTIVITE ENDOCHITINASE.	
INVENTOR(S):	BLAISEAU PIERRE-LOUIS; LEGOUX RICHARDLEGUAY JEAN-JACQUES; SCHNEIDER MICHEL	
PATENT APPLICANT(S):	ELF SANOFI; ELF AQUITAINE STE NALE	
PATENT APPL. COUNTRY:	FR; FR	
LANGUAGE OF FILING:	French	
LANGUAGE OF PUBL.:	French	
DOCUMENT TYPE:	Patent	
PATENT INFO TYPE:	FRB1 PATENT OF INVENTION (SECOND PUBLICATION) (FROM 2,000,000)	
PATENT INFORMATION:		

NUMBER	KIND	DATE

	FR 2681076	B1	19941118
APPLICATION INFO.:	FR 1991-11072	A	19910906
PRIORITY INFO.:	FR 1991-11072	A	19910906

DETDEN . . . continued by chromatography of excLusion on a reticule agarose (column Superose 12 Pharmacia), elution being carried out by a buffer solution of sodium acetate 500 mm of pH 5,2.With each stage, the chitinase is identified by its molecular weight (electrophoresis on polyacrylamide gel to 12,5 % in the presence of SDS-revelation with the money) and its enzymatic activity, measured by the radiochemical method described hereafter using the chitin marked with tritium like substrate (Molano et al.. (1977) Anal. Biochem 83, 648-656).With the exit of the purification, one isolated a protein from apparent molecular weight of 41 _ + 3 kDa which. . . kit of marking of Boehringer MannheimGMBH (ref: 1004 760), used according to the recommendations of the manufacturer. The specific activity obtained is 1 X 10 dpm/ug of ADN.The counterparts on membrane are prehybridees during 1 Hat 65 C in a plug of composition: 6 X SSC;5 X solution ofDenhardt;0,5 % SDS and 100 pg/ml of ADN of sonique salmon sperm. The counterparts on membrane are hybridees with probe 2681076 prepared previously during 16 H in the same plug, then are Lavees during 20 min at 20 C in a plug 2 X SSC;0,1 % SDS, thenduring 40 min in a plug 2 X SSC;0,1 % SDS at 65 C, and finally during 40 min in a plug 0,2 X SSC;0,1 % SDS at 65 C, then driedand autoradiographiees. In short. The plug 20 X SSC contains 175,3 gAL of NaCL;88,2 g/L of sodium citrate and is adjusted with pH 7 by some NacOh 10N drops. The solution 10 X Denhardt contains 1 G of FicoLL 400, 1 G of polyvinylpyrrolidone, 1 G. . .

. . . M, during 5 min. The counterparts are then plunged in a solution of 25 2 XSSC (NaCl 0,30 M, sodium 0,030M citrate).One discusses then the counterparts on membrane with proteinase K (Boehringer Mannheim GMBH) with 100 ug/ml in a solution of composition: Sorting-HCl 10 mm pH 8;EDTA 10 mm;NaCL 50 mm;SDS 0,1 % at a rate of 20 ml per membrane. One incubates during 30 min at 37 C with agitation. The counterparts on membrane are again plunged in a solution of 2 X SSC and the bacterial remains are partially eliminated while rubbing gently with a paper of the mark Kim Wipes. The membranes are then discussed during 5 min in a NacOh solution 0,4 M, then briefly rinsed in a solution of 2 X SSC. One thus obtains, for each box, two counterparts on membrane.the 2681076 filters are put at prehybrider in a plug containing 0,1 % SDS, 6 X SSC, 10 X Denhardt and 100 ug/ml of ADN of sonique and denatured salmon sperm (Sigma). The temperature of prehybridation is of 42°C and the duration of 6 H.Hybridization is carried out at 42 C during 16 H by adding60 ng/ml mixture of the 3 probes marked to peroxidase.The washing of the membranes is ensured in solution X SSC;0,1 % SDS with 22°C during 2 times 5 min, then during min, then by 2 washings of 15 min in the solution 0,1 X SSC + 10 0,1 % SDS with 42°C and finally 3 min in a solution with 2 X SSC with 22°C.The revelation is done using kit ECL of Amersham (ref. RPN2110) according to the protocol of the manufacturer by using the films Xomat AR (Kodak). colonies forwarded a very strong hybridization with 15 the mixture of. . . Ala Gly Val Glu 20 25 30 Lilies Arg the mature protein is the protein of 389 amino acids of a molecular weight close to 42,8 kDa which starts with the sequence aminoterminale (data base determined in section 1. The apparent molecular weight observed approximately 41 _ + _ 3 kDa corresponds, because of the experimental margin of error, with the molecular weight of 42,8 kDa calculated protein deduced from the complementary ADN. This protein has two potential sites of N-glycosylation (stressed on figure

1). Comparison of the peptide sequence (have) to the other already known peptide sequences the comparison carried on the vegetable chitinases of classes I, II and III, defined by Shinshi and Al,. . . then diluted in a plug of charge of following composition:-0,125 M Sorting-HCl pH 6,8 30-4 % dedocylsulfate of sodium-20 % glycerol-0,002 % blue of bromophenol-10 % p-mercaptoethanol then the mixture are carried at 100 C during 10 min. 10 solubilized protein ug are deposited on a gel of electrophoresis of SDS 2681076 polyacrylamide according to the protocol describes by Laemmli (Laemmli, Nature, 227, 1970, 680-685). After electrophoresis, the proteins of the freezing are transferred on a Immobilon membrane (in PVDF) by electrotransfertaccording to the technique from H. Towbin and Al, Proc. 05 Natl.Acad. Sci. The USA, 76, 1979, 4350-4354.The immuno' detection. . .

. . .

min, then centrifuged during 30 min. The base was included in approximately 1 cold ml of ace- tone (+4 C) and centrifuged again 30 min. The base, after being dried, is included in approximately 20 pi of a plug called plug of charge made up of Sorting-HCl 0,125 pH 6,8 SDS 4 %, blue of bromophenol 0,002 %, glycerol 20 %, (3-mercaptoethanol 10 % (according to protocol describes by Laemmli (1970)). The base is solubi- Lise by boiling during 15 min, thenneutralized by adding soda 10 N.The analysis of proteins by electrophoresis in denaturing gel SDS is carried out according to the method described in the section 9d).The profile obtained shows the presence of a supernumeraryWide strip present in the EMY761/pEMR698 stock and absent from the pilot stock (not transformed stock EMY761). This band has a molecular weight ranging between 39 and 46 kDa. The width. . .

. . .

of the expert and in particular described per H. Towbin and Al, Proc. Ntl. Acad. Sci. The USA, 76, 1979, 4350-4354, which includes/understands the following stages:-denaturation by heating with 100° during 10 min in a plug, 15 called plug of charge made up of Sorting HCl 0,125 M pH 6,8, SDS 4 %, bromophenol blue 0,002 %, glycerol 20 %, p-mercapto-ethanol 10 % (according to the protocol describes by Laemmli, the U.K. Laemmli, Nature, 227, 1970, 680-685);-electrophoretic separation of thevarious proteins contained 20 in solubilized according to the protocoldescribed by Laemmli (ref. above);-electrotransfert of the aforesaid proteins contained in the freezing. . .

DETDFR A chaque etape, la chitinase est identifiee par son poids moleculaire electrophorese sur gel de polyacrylamide a 12,5 % en presence de SDS - revelation a l'argent) et son activite enzymatique, mesuree par la methode radiochimique decrite ci-apres utilisant la chitine marquee au tritium comme substrat (Molano et al.

With each stage, the chitinase is identified by its molecular weight (electrophoresis on polyacrylamide gel to 12,5% in the presence of SDS - revelation with the money) and its enzymatic activity, measured by the radiochemical method described hereafter using the chitin marked with tritium like substrate (Molano and al.

A chaque etape, la chitinase est identifiee par son poids moleculaire (electrophorese sur gel de polyacrylamide a 12,5 % en presence de SDS - revelation a l'argent) et son activite enzymatique, mesuree par la methode radiochimique decrite ci-apres utilisant la chitine marquee au tritium comme substrat (Molano et al.

Les repliques sur membrane sont prehybridees pendant I h dans un tampon de composition : 6 x SSC ; 5 x solution de ; 0,5 % SDS et 100

pg/ml d'ADN de sperme de saumon Les repliques sur membrane sont hybridees avec la sonde preparee precedemment pendant 16 h dans le meme tampon, puis sont lavees pendant 20 min a 20 °C dans un tampon 2 x SSC ; 0,1% SDS, puis pendant 40 min dans un tampon 2 x SSC ; 0,1% SDS a 65°C, et enfin pendant 40 min dans un tampon 0,2 x SSC ; 0,1% SDS a 65°C, puis sechees et autoradiographiees. En resume le tampon 20 x SSC contient 175,3 gfl de NaCl ; 88,2 g/l de citrate de sodium et est ajuste a pH 7 par quelques gouttes de NaOH ION. La solution 10 x Denhardt contient 1 g de Ficoll 400, 1 g. . .

in a plug of composition: 6 X SC; 5 X solution of; 0,5% SDS and 100 pg/ml of DNA of salmon sperm the counterparts on membrane are hybridees with the probe prepared previously during 16:00 in the m me plug, then washed then finally then is during 20 min with 20 °C in a plug 2 X SC; 0,1% SDS, during 40 min in a plug 2 X SC; 0,1% SDS with 65°C, and during 40 min in a plug 0,2 X SC; 0,1% SDS with 65°C, dried and autoradiographiees. In short, the plug 20 X SC contains adjusted in Denhardt 1 G of bovine serum albumin for 500 ml of final.

dans un tampon de composition : 6 x SSC ; 5 x solution de ; 0,5 % SDS et 100 pg/ml d'ADN de sperme de saumon Les repliques sur membrane sont hybridees avec la sonde preparee precedemment pendant 16 h dans le m me tampon, puis lavees puis enfin puis sont pendant 20 min a 20 °C dans un tampon 2 x SSC ; 0,1% SDS, pendant 40 min dans un tampon 2 x SSC ; 0,1% SDS a 65°C, et pendant 40 min dans un tampon 0,2 x SSC ; 0,1% SDS a 65°C, sechees et autoradiographiees. En resume, le tampon 20 x SSC contient ajuste a Denhardt 1 g d'albumine serique bovine pour 500 ml de volume final.

On traite ensuite les repliques sur membrane avec de la proteinase K (Boehringer Mannheim GmbH) a 100 pg/ml dans une solution de composition : Tris-HCl 10 mM pH 8 ; EDTA 10 mM ; NaCl mM ; SDS 0,1% a raison de 20 ml par membrane. On incube pendant min a37°C avec agitation. Les repliques sur membrane sont de nouveau plongees dans une solution de 2 x SSC et les debris bacteriens sont partiellement elimines en frottant doucement avec un papier de la marque Kim Wipes. Les membranes. . .

Les filtres sont mis a prehybrider dans un tampon contenant 0,1% SDS, 6 x SSC, 10 x Denhardt et 100 pg/ml d'ADN de sperme de saumon sonique et denature (Sigma). La temperature de prehybridation est de 42 °C et la duree de 6 h.

One treats then the counterparts on membrane with proteinase K (Boehringer Mannheim GMBH) with 100 pg/ml in a solution of composition: Sorting-HCl 10 mm pH 8; EDTA 10 mm; NaCl mm; SDS 0,1% at a rate of 20 ml per membrane. One incubates during min a37°C with agitation. The counterparts on membrane are again plunged in a solution of 2 X SC and the bacterial remains are partially eliminated while rubbing gently with a paper of the mark Kim Wipes. The membranes. . .

On traite ensuite les repliques sur membrane avec de la proteinase K (Boehringer Mannheim GmbH) a 100 pg/ml dans une solution de composition : Tris-HCl 10 mM pH 8 ; EDTA 10 mM ; NaCl mM ; SDS 0,1% a raison de 20 ml par membrane. On incube pendant min a37°C avec agitation. Les repliques sur membrane sont de nouveau plongees dans une solution de 2 x SSC et les debris bacteriens sont partiellement elimines en frottant doucement avec un papier de la marque Kim Wipes. Les

membranes. . .

The filters are put at prehybrider in a plug containing 0,1% SDS , 6 X SC, 10 X Denhardt and 100 pg/ml of DNA of sonic and denatured salmon sperm (Sigma). The temperature of prehybridation is of 42°C and the duration of 6:00

Les filtres sont mis a prehybrider dans un tampon contenant 0,1% SDS, 6 x SSC, 10 x Denhardt et 100 pg/ml d'ADN de sperme de saumon sonique et denature (Sigma). La temperature de prehybridation est de 42°C et la duree de 6 h.

Le lavage des membranes est assure dans la solution 2 x SSC ; 0,1% SDS a 22°C pendant 2 fois 5 min, puis pendant min, puis par 2 lavages de 15 min dans la solution 0,1 x SSC + 0,1% SDS a 42 °C et enfin 3 min dans une solution a 2 x SSC a22°C.

The washing of the membranes is ensured in the solution 2 X SC; 0,1% SDS with 22°C during 2 times 5 min, then during min, then by 2 washings of 15 min in the solution 0,1 X SC + 0,1% SDS with 42°C and finally 3 min in a solution with 2 X SC with 22°C.

Le lavage des membranes est assure dans la solution 2 x SSC ; 0,1% SDS a 22°C pendant 2 fois 5 min, puis pendant min, puis par 2 lavages de 15 min dans la solution 0,1 x SSC + 0,1% SDS a 42°C et enfin 3 min dans une solution a 2 x SSC a 22°C.

. . .
Ala Thr Pro Island Ser Ser Glu Went Gly Val Lime Lily Arg the mature protein is the protein of 389 amino-acids of a molecular weight close to 42,8 kDa which starts with the sequence aminoterminale (bl) given in section 1. The apparent molecular weight observed approximately 41 + 3 kDa corresponds, taking into account the experimental margin of error, with the molecular weight of 42,8 kDa calculated protein deduced from the complementary DNA. This protein has two potential sites of Nglycosylation (underlined on figure 1).

- 0,125 M Tris-HCl pli 6,8 - 4 % dedocylsuLfate de sodium -20 % glycerol - 0,002 % bleu de bromophenol - 10 % -mercaptoethanol puis le melange est porte a I00 °C pendant 10 min. 10 pg de proteines solubilisees sont deposes sur un gel d'electrophorese de SDS polyacrylamide selon le protocole decrit par Laemmli (Laemmli, Nature, 227, 1970, 680-685). Apres electrophorese, les proteines du gel sont transferees sur une membrane Immobilon (en PVDF) par eLectrotransfert selon la technique de H. Towbin et al., Proc.

- 0,125 M Sorting-HCl fold 6,8 - 4% dedocylsulfate of sodium - 20% glycerol - 0,002% blue of bromophenol - 10% - mercaptoethanol then the mixture are carried to 100 °C during 10 min. 10 solubilized protein pg are deposited on a gel of electrophoresis of SDS polyacrylamide according to the protocol describes by Laemmli (Laemmli, Nature, 227,1970,680-685). After electrophoresis, the proteins of freezing are transferred on a Immobilon membrane (in PVDF) by electrotransfert according to the technique from H. Towbin and Al, Proc.

- 0,125 M Tris-HCl pli 6,8 - 4 % dedocylsulfate de sodium - 20 % glycerol - 0,002 % bleu de bromophenol - 10 % -mercaptoethanol puis le melange est porte a 100 °C pendant 10 min. 10 pg de proteines solubilisees sont deposes sur un gel d'electrophorese de SDS polyacrylamide selon le protocole decrit par Laemmli (Laemmli, Nature,

227, 1970, 680-685). Apres electrophorese, les proteines du gel sont transferees sur une membrane Immobilon (en PVDF) par electrotransfert selon la technique de H. Towbin et al., Proc.

. . .

pendant 30 min, puis centrifuge pendant 30 min. Le culot a ete repris dans environ 1 ml d'acetone froid (+4°C) et de nouveau centrifuge 30 min. Le culot, apres avoir ete seche, est repris dans environ 20 c1 d'un tampon denomme tampon de charge constitue de Tris-HCl 0,125 pH 6,8 SDS 4 %, bleu de bromophenol 0,002 %, glycerol 20 %, -mercaptoethanol 10 % (selon protocole decrit par Laemmli (1970)). Le culot est solubilise par ebullition pendant 15 min, puis neutralise en ajoutant de la soude 10 N.

. . .

during 30 min, then centrifuged during 30 min. the base was included in approximately 1 cold ml of acetone (+4°C) and centrifuged 30 again min. the base, after being dried, is included in approximately 20 pl d' a plug called plug of load made up of Sorting-HCl 0,125 pH 6,8 SDS 4%, blue of bromophenol 0,002%, glycerol 20%, C-mercaptoethanol 10% (according to protocol describes by Laemmli (1970)). The base is solubilized by boiling during 15 min, then neutralized by adding soda 10 N.

. . .

pendant 30 min, puis centrifuge pendant 30 min. Le culot a ete repris dans environ 1 ml d'acetone froid (+4°C) et de nouveau centrifuge 30 min. Le culot, apres avoir ete seche, est repris dans environ 20 pl d'un tampon denomme tampon de charge constitue de Tris-HCl 0,125 pH 6,8 SDS 4 %, bleu de bromophenol 0,002 %, glycerol 20 %, C-mercaptoethanol 10 % (selon protocole decrit par Laemmli (1970)). Le culot est solubilise par ebullition pendant 15 min, puis neutralise en ajoutant de la soude 10 N.

L'analyse des proteines par electrophorese en gel SDS denaturant est realisee selon la methode decrite dans la section 9d).

The analysis of proteins by electrophoresis in denaturing gel SDS is carried out according to the method described in the section 9d).

L'analyse des proteines par electrophorese en gel SDS denaturant est realisee selon la methode decrite dans la section 9d).

- denaturation by heating with 100° during 10 min in a plug, called plug of load made up of Sorting HCI 0,125 M fold 6,8, SDS 4%, blue of bromophenol 0,002%, glycerol 20%, - mercaptoethanol 10% (according to the protocol describes by Laemmli, U.K.

- denaturation par chauffage a 100° pendant 10 min dans un tampon, denomme tampon de charge constitue de Tris HCI 0,125 M pli 6,8, SDS 4 %, bleu de bromophenol 0,002 %, glycerol 20 %, -mercaptoethanol 10 % (selon le protocole decrit par Laemmli, U.K.

- denaturation par chauffage a 100° pendant 10 min dans un tampon, denomme tampon de charge constitue de Tris HCI 0,125 M pli 6,8, SDS 4 %, bleu de bromophenol 0,002 %, glycerol 20 %, -mercaptoethanol 10 % (selon le protocole decrit par Laemmli, U.K.

FILE 'HOME' ENTERED AT 15:27:47 ON 15 JUN 2011

SEARCH HISTORY

```
=> d his nofile

     (FILE 'HOME' ENTERED AT 14:28:50 ON 15 JUN 2011)

     FILE 'CAPLUS' ENTERED AT 14:29:06 ON 15 JUN 2011
L1         12367 SEA SPE=ON  ABB=ON  SMITH G?/AU
L2            19 SEA SPE=ON  ABB=ON  KNELL J?/AU
L3            16 SEA SPE=ON  ABB=ON  VOZNESENSKY A?/AU
L4             2 SEA SPE=ON  ABB=ON  L1 AND L2 AND L3
                 D SCA
                 E POLYCYTHEMI/BI
L5           687 SEA SPE=ON  ABB=ON  POLYCYTHEMIC/BI
L6        455651 SEA SPE=ON  ABB=ON  MICE/OBI OR MOUSE/OBI OR MURINE/OBI
L7        924266 SEA SPE=ON  ABB=ON  (MICE OR MOUSE OR MURINE)/BI
L8           416 SEA SPE=ON  ABB=ON  L5 AND L7
L9           360 SEA SPE=ON  ABB=ON  L5(3A) L7
L10        72030 SEA SPE=ON  ABB=ON  ?HYPOXI?/BI
L11          107 SEA SPE=ON  ABB=ON  L9(3A)L10
                 D KWIC
                 D KWIC 10
L12           73 SEA SPE=ON  ABB=ON  (EXHYPOXIC OR EX(A)HYPOXIC)/BI
L13           64 SEA SPE=ON  ABB=ON  L9(3A)L12
L14           63 SEA SPE=ON  ABB=ON  L12(W)L5(W)L7
L15            1 SEA SPE=ON  ABB=ON  L13 NOT L14
                 D KWIC
L16           65 SEA SPE=ON  ABB=ON  L5(A)L12
L17           64 SEA SPE=ON  ABB=ON  L16(W)L7

     INDEX '1MOBILITY, 2MOBILITY, ADISCTI, AEROSPACE, AGRICOLA, ALUMINIUM,
     ANABSTR, ANTE, APOLLIT, AQUALINE, AQUASCI, BABS, BIBLIODATA, BIOENG,
     BIOSIS, BIOTECHABS, BIOTECHDS, BIOTECHNO, CABA, CAPLUS, CASREACT, CBNB,
     CEABA-VTB, CERAB, CHEMINFORMRX, CHEMSAFE, ...' ENTERED AT 14:45:49 ON 15
     JUN 2011
                 SEA (EX HYPOXIC OR EXHYPOXIC) AND (POLYCYTHEMIC OR POLY CYTHEMI
               ---------
                 1   FILE ANABSTR
                 1   FILE BIOENG
                56   FILE BIOSIS
                10   FILE BIOTECHNO
                 2   FILE CABA
                 4   FILE CAPLUS
                 1   FILE CONFSCI
                 3   FILE DDFB
                 4   FILE DDFU
                 5   FILE DISSABS
                 3   FILE DRUGB
                 8   FILE DRUGU
                79   FILE EMBASE
                 4   FILE ENERGY
                28   FILE EPFULL
                 3   FILE ESBIOBASE
                 2   FILE FRFULL
                 1   FILE IFIPAT
                 3   FILE INIS
                 1   FILE IPA
                 4   FILE LIFESCI
                53   FILE MEDLINE
                 3   FILE NTIS
```

```
                7   FILE PASCAL
               49   FILE PCTFULL
               12   FILE SCISEARCH
               24   FILE TOXCENTER
               71   FILE USPATFULL
               15   FILE USPAT2
                2   FILE WPIDS
                2   FILE WPINDEX
L18              QUE SPE=ON  ABB=ON  (EX HYPOXIC OR EXHYPOXIC) AND (POLYCYTHEMIC
                  OR POLY CYTHEMIC)
                 ---------
                 D RANK

     FILE 'STNGUIDE' ENTERED AT 14:46:54 ON 15 JUN 2011

     FILE 'MEDLINE, DRUGU, DRUGB, PASCAL, BIOTECHNO, WPIX, IPA, BIOSIS,
     LIFESCI, CONFSCI, ESBIOBASE, NTIS, DISSABS, EMBASE, BIOENG, ANABSTR,
     SCISEARCH' ENTERED AT 14:50:07 ON 15 JUN 2011
L19          291 SEA SPE=ON  ABB=ON  (EX HYPOXIC OR EXHYPOXIC)
L20         4165 SEA SPE=ON  ABB=ON  (POLYCYTHEMIC OR POLY CYTHEMIC)
L21      6672721 SEA SPE=ON  ABB=ON  MOUSE OR MICE OR MURINE
L22          248 SEA SPE=ON  ABB=ON  L19 AND L20 AND L21
L23          242 SEA SPE=ON  ABB=ON  L19(3A) L20(3A) L21

     FILE 'STNGUIDE' ENTERED AT 14:51:16 ON 15 JUN 2011

     FILE 'MEDLINE, DRUGU, DRUGB, PASCAL, BIOTECHNO, WPIX, IPA, BIOSIS,
     LIFESCI, CONFSCI, ESBIOBASE, NTIS, DISSABS, EMBASE, BIOENG, ANABSTR,
     SCISEARCH' ENTERED AT 14:51:48 ON 15 JUN 2011
                 D QUE L22
L24            2 SEA SPE=ON  ABB=ON  L22 AND PATENT/DT
L25            0 SEA SPE=ON  ABB=ON  L22 AND (REVIEW/DT OR GENERAL REVIEW/DT)
L26          246 SEA SPE=ON  ABB=ON  L22 NOT L24
L27          221 SEA SPE=ON  ABB=ON  L26 AND PY<1999
L28            2 SEA SPE=ON  ABB=ON  L24 AND (PD<19981008 OR AD<19981008 OR
                 PRD<19981008)
L29          223 SEA SPE=ON  ABB=ON  (L27 OR L28)
L30       160030 SEA SPE=ON  ABB=ON  EPO OR ERYTHROPOIETIN
L31        83860 SEA SPE=ON  ABB=ON  BACULOVIR?
L32        61112 SEA SPE=ON  ABB=ON  INSECT#(2A) CELL#
L33          203 SEA SPE=ON  ABB=ON  L29 AND L30
L34            0 SEA SPE=ON  ABB=ON  L29 AND L30 AND (L31 OR L32)
L35      2239817 SEA SPE=ON  ABB=ON  RECOMB?
L36        48288 SEA SPE=ON  ABB=ON  L30(3A) L35
L37            9 SEA SPE=ON  ABB=ON  L29 AND L36
L38           15 SEA SPE=ON  ABB=ON  L29 AND L30 AND L35
L39           99 DUP REM L29 (124 DUPLICATES REMOVED)
                       ANSWERS '1-49' FROM FILE MEDLINE
                       ANSWERS '50-54' FROM FILE DRUGU
                       ANSWERS '55-57' FROM FILE DRUGB
                       ANSWER '58' FROM FILE PASCAL
                       ANSWERS '59-60' FROM FILE BIOTECHNO
                       ANSWERS '61-62' FROM FILE WPIX
                       ANSWER '63' FROM FILE IPA
                       ANSWERS '64-74' FROM FILE BIOSIS
                       ANSWER '75' FROM FILE CONFSCI
                       ANSWERS '76-77' FROM FILE NTIS
                       ANSWERS '78-82' FROM FILE DISSABS
                       ANSWERS '83-98' FROM FILE EMBASE
                       ANSWER '99' FROM FILE ANABSTR
```

```
     FILE 'CAPLUS' ENTERED AT 14:58:11 ON 15 JUN 2011
                D QUE L13
                D SCA L4

     FILE 'REGISTRY' ENTERED AT 14:58:55 ON 15 JUN 2011
L40             1 SEA SPE=ON  ABB=ON  11096-26-7
                  E ERYTHROPOIETIN/CN
L41             1 SEA SPE=ON  ABB=ON  ERYTHROPOIETIN/CN
                  D SCA
                  D SCA L40
L42             1 SEA SPE=ON  ABB=ON  (L40 OR L41)

     FILE 'CAPLUS' ENTERED AT 14:59:33 ON 15 JUN 2011
L43         15109 SEA SPE=ON  ABB=ON  L41
L44             1 SEA SPE=ON  ABB=ON  L13 AND PATENT/DT
L45             1 SEA SPE=ON  ABB=ON  L13 AND REVIEW/DT
L46            63 SEA SPE=ON  ABB=ON  L13 NOT L44
L47            59 SEA SPE=ON  ABB=ON  L46 AND PY<1999
L48             1 SEA SPE=ON  ABB=ON  L44 AND (PD<19981008 OR AD<19981008 OR
                  PRD<19981008)
L49            60 SEA SPE=ON  ABB=ON  (L47 OR L48 OR L45)
L50            52 SEA SPE=ON  ABB=ON  L43 AND L49
L51        225465 SEA SPE=ON  ABB=ON  RECOMB?/OBI
L52          1789 SEA SPE=ON  ABB=ON  L43(L)L51
L53             2 SEA SPE=ON  ABB=ON  L49 AND L52
L54          8016 SEA SPE=ON  ABB=ON  BACULOVIR?/OBI
L55         11949 SEA SPE=ON  ABB=ON  (INSECT#(2A)CELL#)/BI
L56             0 SEA SPE=ON  ABB=ON  L49 AND (L54 OR L55)
                  D QUE
                  D QUE L50
L57             0 SEA SPE=ON  ABB=ON  L50 AND (L54 OR L55)
                  D SCA L4
L58          1228 SEA SPE=ON  ABB=ON  INSECT#/OBI(L)TISSUE/OBI
L59             0 SEA SPE=ON  ABB=ON  L50 AND L58
L60        579127 SEA SPE=ON  ABB=ON  VIVO/BI
L61            11 SEA SPE=ON  ABB=ON  L50 AND L60

     FILE 'MEDLINE, DRUGU, DRUGB, PASCAL, BIOTECHNO, WPIX, IPA, BIOSIS,
     LIFESCI, CONFSCI, ESBIOBASE, NTIS, DISSABS, EMBASE, BIOENG, ANABSTR,
     SCISEARCH' ENTERED AT 15:09:41 ON 15 JUN 2011
L62       4344294 SEA SPE=ON  ABB=ON  VIVO
                  D QUE L33
L63            41 SEA SPE=ON  ABB=ON  L33 AND L62
L64            23 DUP REM L63 (18 DUPLICATES REMOVED)
                        ANSWERS '1-8' FROM FILE MEDLINE
                        ANSWERS '9-14' FROM FILE DRUGU
                        ANSWER '15' FROM FILE PASCAL
                        ANSWER '16' FROM FILE WPIX
                        ANSWER '17' FROM FILE BIOSIS
                        ANSWER '18' FROM FILE DISSABS
                        ANSWERS '19-23' FROM FILE EMBASE

     FILE 'STNGUIDE' ENTERED AT 15:10:20 ON 15 JUN 2011

     FILE 'MEDLINE, DRUGU, DRUGB, PASCAL, BIOTECHNO, WPIX, IPA, BIOSIS,
     LIFESCI, CONFSCI, ESBIOBASE, NTIS, DISSABS, EMBASE, BIOENG, ANABSTR,
     SCISEARCH' ENTERED AT 15:11:18 ON 15 JUN 2011
                D QUE L34
                D QUE L38
```

```
                D QUE L63
L65           52 SEA SPE=ON  ABB=ON  (L38 OR L63)

     FILE 'CAPLUS' ENTERED AT 15:11:22 ON 15 JUN 2011
                D QUE L53
                D QUE L61
                D QUE L57
                D QUE L59
L66           13 SEA SPE=ON  ABB=ON  (L53 OR L61)

     FILE 'MEDLINE, DRUGU, PASCAL, BIOTECHNO, WPIX, BIOSIS, DISSABS, EMBASE,
     ANABSTR, SCISEARCH, CAPLUS' ENTERED AT 15:11:33 ON 15 JUN 2011
L67           33 DUP REM L65 L66 (32 DUPLICATES REMOVED)
                     ANSWERS '1-11' FROM FILE MEDLINE
                     ANSWERS '12-18' FROM FILE DRUGU
                     ANSWER '19' FROM FILE PASCAL
                     ANSWER '20' FROM FILE BIOTECHNO
                     ANSWER '21' FROM FILE WPIX
                     ANSWER '22' FROM FILE BIOSIS
                     ANSWER '23' FROM FILE DISSABS
                     ANSWERS '24-28' FROM FILE EMBASE
                     ANSWER '29' FROM FILE ANABSTR
                     ANSWERS '30-33' FROM FILE CAPLUS
                D IALL 1-20
                D IFULL 21
                D IALL 22-29
                D IBIB AB HITIND 30-33

     FILE 'HOME' ENTERED AT 15:12:17 ON 15 JUN 2011

     INDEX '1MOBILITY, 2MOBILITY, ADISCTI, AEROSPACE, AGRICOLA, ALUMINIUM,
     ANABSTR, ANTE, APOLLIT, AQUALINE, AQUASCI, BABS, BIBLIODATA, BIOENG,
     BIOSIS, BIOTECHABS, BIOTECHDS, BIOTECHNO, CABA, CAPLUS, CASREACT, CBNB,
     CEABA-VTB, CERAB, CHEMINFORMRX, CHEMSAFE, ...' ENTERED AT 15:12:46 ON 15
     JUN 2011
                SEA MARGIN#(1W)ERROR AN
               ---------
              11   FILE 1MOBILITY
                SEA MARGIN#(1W)ERROR AND (SDS OR SODIUM DODECYL? OR SODIUMDODEC
               ---------
              74   FILE EPFULL
               9   FILE FRFULL
               7   FILE GBFULL
             317   FILE PCTFULL
             483   FILE USPATFULL
             109   FILE USPAT2
L68             QUE SPE=ON  ABB=ON  MARGIN#(1W)ERROR AND (SDS OR SODIUM
                DODECYL? OR SODIUMDODECYL?)
               ---------

     FILE 'USPATFULL, PCTFULL, USPAT2, EPFULL, FRFULL, GBFULL' ENTERED AT
     15:16:22 ON 15 JUN 2011
L69        18198 SEA SPE=ON  ABB=ON  MARGIN#(1W) ERROR
L70       317803 SEA SPE=ON  ABB=ON  SDSPAGE OR SDS OR SODIUM DODECYL? OR
                 SODIUMDODECYL?
L71            0 SEA SPE=ON  ABB=ON  L69(5A) L70
L72          156 SEA SPE=ON  ABB=ON  L69(S) L70
                 D KWIC 1 50 100 150
L73      1073173 SEA SPE=ON  ABB=ON  MOLECULAR WEIGHT
L74       433002 SEA SPE=ON  ABB=ON  MW OR M(W) W
```

```
L75          21 SEA SPE=ON  ABB=ON  L69(8A) (L73 OR L74)
L76           4 SEA SPE=ON  ABB=ON  L75 AND L70
                D KWIC 1-4
L77      243292 SEA SPE=ON  ABB=ON  KDA OR KILODALTON# OR DALTON#
L78           8 SEA SPE=ON  ABB=ON  L69(8A) L77 AND L70
L79      251969 SEA SPE=ON  ABB=ON  FRACTIONAT?
L80           0 SEA SPE=ON  ABB=ON  L69(8A) L79 AND L70
L81           5 SEA SPE=ON  ABB=ON  L69(S) L79 AND L70
                D KWIC 1-5

     FILE 'STNGUIDE' ENTERED AT 15:25:48 ON 15 JUN 2011

     FILE 'USPATFULL, PCTFULL, USPAT2, EPFULL, FRFULL, GBFULL' ENTERED AT
     15:26:34 ON 15 JUN 2011
                D QUE L76
                D QUE L78
                D QUE L71
                D QUE L80
L82           9 SEA SPE=ON  ABB=ON  (L76 OR L78)
L83           9 DUP REM L82 (0 DUPLICATES REMOVED)
                      ANSWERS '1-2' FROM FILE USPATFULL
                      ANSWERS '3-6' FROM FILE PCTFULL
                      ANSWER '7' FROM FILE EPFULL
                      ANSWERS '8-9' FROM FILE FRFULL
                D IBIB AB KWIC 1-9

     FILE 'HOME' ENTERED AT 15:27:47 ON 15 JUN 2011

=>
```